SOME INDIAN GREEN AND BROWN ALGAE ESPECIALLY FROM THE SHORES OF THE PRESIDENCY OF BOMBAY—III.[1]

BY

F. BOERGESEN.

Fam. Valoniaceæ.[2]

Valonia utricularis (Roth.) Ag.

Agardh, C., Spec. Alg., vol. I, 1821, p. 431. *Kuckuck, P.*, Über den Bau und die Fortpflanzung von Halicystis Aresch. und Valonia Ginn., Bot. Zeit. 1907. *Boergesen, F.*, Mar. Algæ D. W. I., vol. I, p. 30, fig. 17–18.

The specimens form low tufts on the rocks. The vesicles are often rather much ramified and intermingled forming low cushions. The form found seems to come near forma CRUSTACEA Kuck.

India : Okha Port.

Geogr. Distrib. In most warm seas.

Fam. Boodleaceæ.

Cladophoropsis Zollingeri (Kütz.) Boergs.

Boergesen, F., Contributions à la connaissance du genre Siphonocladus Schmitz. (Oversigt kgl. danske Videnskab. Selskabs Forhandlinger, 1905, no. 3, p. 288).

---

[1] Continued from the Journal of the Indian Botanical Society, Vol. IX, p. 151 and Vol. XI. p. 51. A corresponding series of papers on the Indian Rhodophyceæ is published in the Kew Bulletin, part I in 1931, No. 1, and part II in 1932, No. 3.

[2] Regarding the arrangement of the families and genera in the group *Siphonocladales* compare my remarks in "Marine Algæ from the Canary Islands", I, Chlorophyceae, in Kgl. Danske Videnskabernes Selskab. Biologiske Meddelelser, Vol. V, 3. København 1925.

CLADOPHORA AEGAGROPILA Zollingeri Kütz., Spec. Alg. p. 415; Tab. Phycolog., vol. IV, tab. 64, fig. II.

SIPHONOCLADUS ZOLLINGERI (Kütz.) Bornet in Mission scientif. du Cap Horn., t. v., Botanique, p. 22. *De Toni*, Sylloge Alg., vol. I, p. 359.

The plant forms dense rather dark-green tufts. The filaments of which the thallus consists are very irregularly ramified (Fig. 1), the
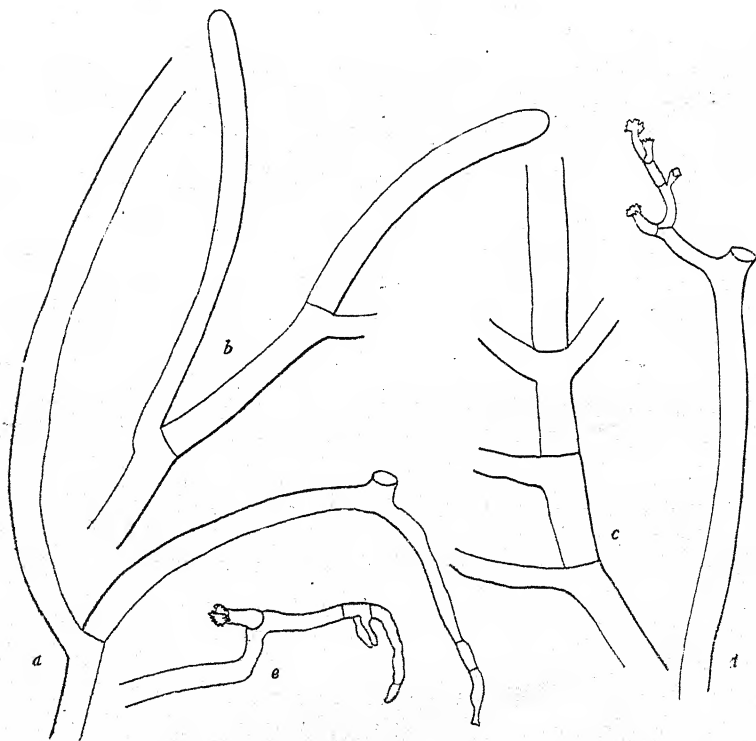

Fig. 1. **Cladophoropsis Zollingeri** (Kütz.) Boergs a, b, c, showing different kind of ramification; a, d, e, filaments with rhizoids and tenacula.   × 22.

branches issuing at shorter or longer intervals, sometimes scattered occasionally more or less uniseriate and rarely subopposite as well. Cross-walls occur very irregularly, the cells forming sometimes very long utricles, sometimes short ones only a few times longer than the breadth of the filaments. The breadth of the filaments is rather variable; most of the filaments are about 150 μ thick, but filaments less than 100 μ or more than 200 μ are often found. The apex of the filaments is obtuse, and near it the filaments are often a little thicker.

At the base the filaments are provided with rhizoids and short tenacula (Fig. 1, d, e) by means of which they are fastened to the substratum and neighbouring filaments.

When I refer this plant to CLADOPHOROPSIS ZOLLINGERI it is because its dimensions seem to agree very well with those of this species, and because on the whole my plant agrees quite well with it ; but I wish to point out that I have not seen any original specimen of this species.

The plant forms dense tufts on the rock in the lower littoral zone in somewhat sheltered places.

India : Bombay in the Arabian Sea.

Geogr. Distrib. Malayan Archipelago.

Fam. Siphonocladiaceæ.
Struvea delicatula Kütz.

Kützing, Tabulæ Phycologicæ, vol. 16, tab. 2.

In earlier papers (cf. Mar. Alg. D.W.I. vol. I, p. 54, fig. 39) I considered *Harvey's* CLADOPHORA ? ANASTOMOSANS and *Kützing's* plant as the same species. Meanwhile by the kind permission of Mr. *J. Ramsbottom,* Keeper of Botany, British Museum, I was able to see a slide of *Harvey's* STRUVEA ANASTOMOSANS, and on comparing it with specimens of STRUVEA DELICATULA, I have come to the conclusion that these are different species. *Harvey's* figure in " Phycologia Australica", Pl. 101, gives a really good representation of the Australian plant. As the most marked differences between *Harvey's* plant and *Kützing's* I shall point out its larger size and its bigger leaf-like part, which is more openly ramified on account of the more distantly placed branchlets. To this must be added the much longer cells in the main stem and branches ; and finally *Harvey's* plant has quite thin walls.

From India I have seen a few specimens of STRUVEA DELICATULA which were given to me by Mr. *S. C. Dixit* and a few others which I gathered myself at Bandra seaface near Bombay. I have compared these specimens with plants from the West Indies and found that they quite agree with my figures and description (l.c.) of it.

India : Bombay, Colaba (legit et dedit Mr. *S. C. Dixit*) ; Bandra seaface.

Geogr. Distrib. Found in most warm seas.

Struvea tuticorinensis Boergs. sp. nov.

STRUVEA cæspites, formans ca 3½ cm alta, e stipitibus cylindricis simplicibus, ad basem transverse annulatis, superne levibus et flabellis

ovalibus, reticulatis composita. Flabellum ca 1½ cm, longum et 1 cm latum in ætate juniori ovalem in adultiori magis irregulariter formatum, costatum, costa e cellulis 500–800 μ longis et 250 μ latis composita. Rami flabelli oppositi, pluries pinnatis, apicibus ramulorum concrescentibus. Plate I et Fig. 2.

The basal part of the plant consists of very much ramified rhizoids by means of which the plant is fixed to the substratum, shells, stones, etc. From this base the erect stems arise. When young they are almost cylindrical bodies a little slender at their base getting gradually thicker upwards and tapering again near the summit into the upper obtuse end. For instance such a young stem at its base is about 330 μ thick and at its thickest about 1,000 μ; its length is 1½ cm. and it is composed of a single cell only. When it reaches the stage of development when the leaflike part of the thallus is going to be formed, some annular corrugations (5–6) become visible at the base of the stem (Fig. 2), and at the same time a row of cells is formed at its upper end. The material in spirits was, I am sorry to say, very bad. It contained only some very young undivided stems and bits of old half-decayed thalli; but to judge from the dried material the development of the leaflike part of the thallus takes place in quite the same way as in STRUVEA ELEGANS as described by me [1]. From the upper end of the cells formed at the apex of the stem two opposite branches grow out at both sides from each of the cells, all the branches lying in the same plane. When these branches have reached some length, they are divided into a number of cells by segregative cell-division and from the upper end of these cells two new opposite branches grow out again (Fig. 2). This process may be repeated a few times more, the last ramifications being issued often more irregularly. The upper ends of the short branchlets become firmly fixed to the neighbouring filaments.

The basal cell of the leaf-like part of the thallus is the longest, about 2 mm., whereas the other cells in the main axis in the leaf reach only a length of about 500–600μ, and their breadth is about 250μ. The height of the plant is a little more than 3 cm; the length of the leaf-like part about 1½ cm and the breadth a little more than 1 cm.

As mentioned above the plant is very much like the West Indian STRUVEA ELEGANS, but on the whole it is much smaller, almost half as small. On account of this and its geographical distribution, I think it most appropriate to consider it a new species.

The plant was dredged at a depth of about 40 feet.

South India : Tuticorin. BOERGESEN no. 5673 (type).

[1] *Boergesen, F.*, Marine Algæ of the Danish West Indies, vol. I, p. 51 figs. 37–38, Copenhagen 1913.

Chamaedoris auriculata Boergs. sp. nov.

Thallus ca 6 cm. altus, e stipite tubuloso, transverse annulato, in superiori parte 1–2 cellulas continente, et capitulo excentrico,
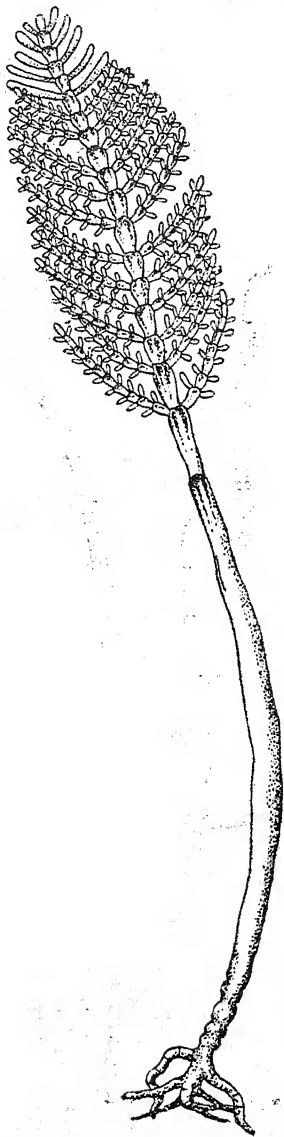

Fig. 2. **Struvea tuticorinensis** Boergs sp. nov. × 5.

auriculato, ad 1½ cm longo compositus. Capitulum e filamentis, in 2–3 verticillas ordinatis, subdichotome ramosis, ca 125 μ latis, inter se tenaculis adfixis compositum est.

This fine plant (Fig. 3) forms more or less dense tufts upon the rocks to which it is fixed by means of ramified rhizoids. The tufts have as a rule a height of about 6 cm; but some of the specimens may
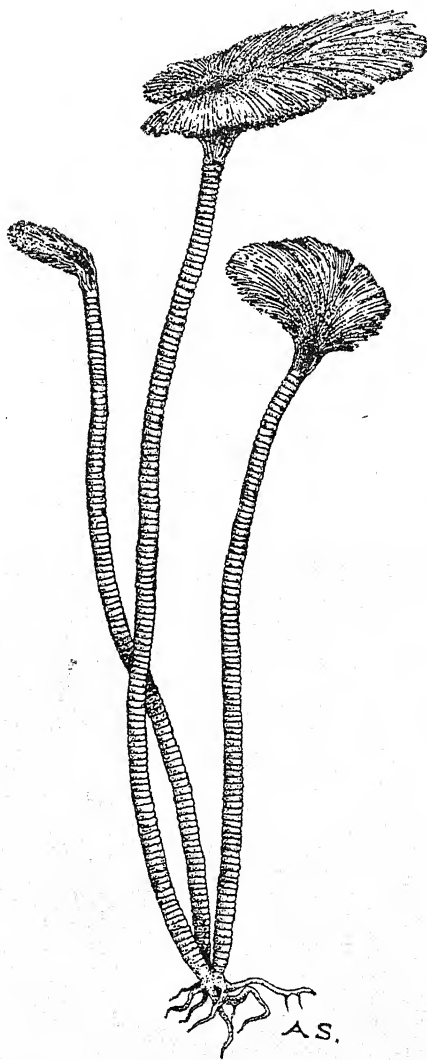

Fig. 3. **Chamædoris auriculata** Boergs. sp. now. Habit of the plant × 2½.

reach a length of up to 8–9 cm. The stipe is annularly constricted from the base to the beginning of the capitulum. Generally the stipe is unbranched, but occasionally it is branched, the branching appearing to be due as a rule to some damage.

The development of the thallus is very similar to that of
CHAMÆDORIS PENICULUM as described by me in "Mar. Alg. D.W.I.",
vol. I, p. 56, figs. 40–43, but owing to the scantiness of the material
available I have not been able to follow it in all its details.   The stipe
is at first a cylindrical tube which gradually becomes annularly
constricted from the base upwards.   When it has reached its normal
size, its upper end swells somewhat and one to two cells are formed.
When two cells are formed, the lower cell is larger than the upper
(Fig. 4a).   Then the formation of the capitulum begins.   From the
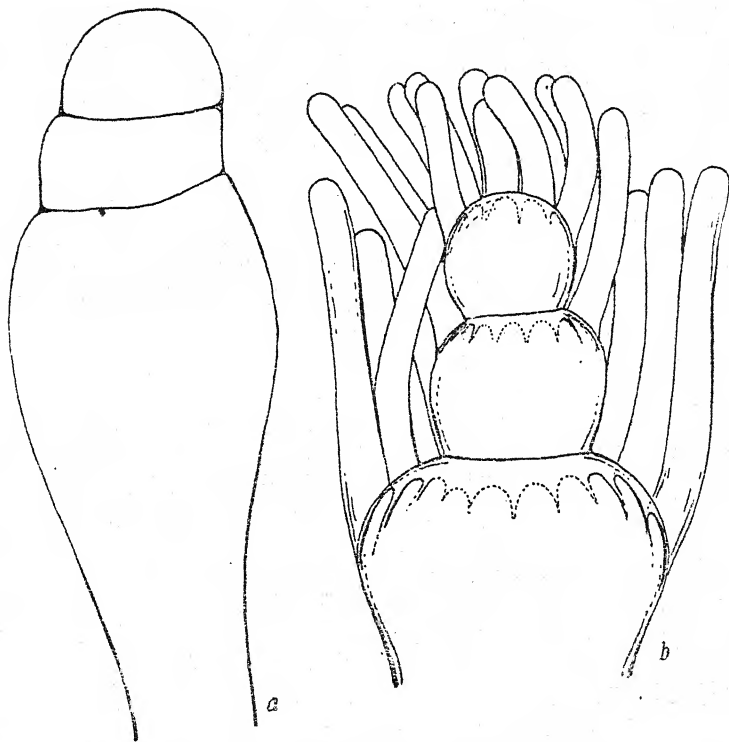upper end of the swollen part of the stem a whorl of filaments is

Fig. 4. **Chamædoris auriculata.** Boergs. a, upper end of a filament with two
cells; b, longitudinal section of a young capitulum.   × 30.

given out surrounding the base of the smaller cell above, and in a
similar way whorls of filaments are given off from the upper end of
this cell and from the one above, if two are present (Fig. 4b).   At the
same time the upper end of the stipe becomes somewhat compressed.
The lowermost whorl consists of about 20 filaments, while the two
above have decreasing numbers of filaments.   The filaments grow out

eccentrically to one side forming the flat, auriculate capitulum (Fig. 3). The filaments are repeatedly subdichotomously ramified (Fig. 5a) and kept together by means of tenacula (Fig. 5b). The diameter of the filaments is about 125 μ.
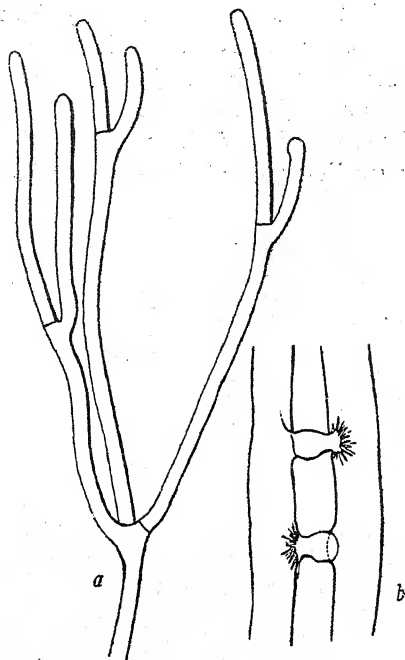

Fig. 5. **Chamædoris auriculata** Boergs. a, upper ends of the filaments composing the capitulum. b, parts of filaments attached to each other by tenacula. a, × 30; b, × 65.

It is very interesting that this genus hitherto considered to be monotypic now comprises 3 species, namely, the long known West Indian species, CHAMÆDORIS PENICULUM (Sol.) Kuntze, one Japanese species, CHAMÆDORIS ORIENTALIS Okamura[1], and the Indian one described here. The Indian plant is closely related to CHAMÆDORIS PENICULUM, but is much smaller, and its capitulum does not reach the height of the West Indian Plant and is easily distinguished from the former species by its thin, oblique, eccentric, rather flat, auriculate capitulum[2] in which only one or two central cells are formed at the top of the stipe.

---

[1] *Okamura, K.*. On the marine Algae from Kôtôsho (Botel Tobago). Reprinted from the Bulletin of the Biogeographical Society of Japan, 1931, vol. 2, p. 98, pl. 10.

[2] The shape of the thallus reminds one very much that of the fungus, HYDNUM AURISCALPIUM L.

The plant was found in the littoral zone in a very exposed locality and formed together with other small algæ dense tufts upon the rocks.

India : Dwarka, *Boergesen* No. 5447 (type).

Fam. Fucaceae.

Cystophyllum muricatum (Turn.) J. Ag.

Agardh, J., Spec. Alg., vol. I, p. 231. *Harvey,* Phycologia Austral. vol. III, pl. 139.

FUCUS MURICATUS Turner, Fuci, p. 107, tab. 112.

CYSTOSEIRA MURICATA Ag., Icones ineditæ, Nova edit., Lund 1846, tab. XII.

CYSTOSEIRA TRINODIS Ag., Icones ineditæ, Holmiæ 1821, tab. XII.

SIROPHYSALIS MURICATA Kütz., Phycol. gener., p. 368; Spec. Alg., p. 602; Tab. phycol., vol. X, tab. 55.

The specimens which I refer to this plant seem to agree quite well with *J. Agardh's* description and with the quoted figures. The plant reaches a height of about 50 cm or perhaps more. It has a rough stem, the roughness being due to numerous short processes which, as strikingly described by *Turner*, remind one of "rudiments of leaves or branches, which give the plant a singularly muricated appearance." The basal leaves, present only in a few specimens, are linear with entire margin, about 4–5 cm long and 3 mm broad with more or less rounded apices and a row of cryptostomes on both sides of the midrib. The vesicles are nearly all of the same size, oval with cryptostomes scattered over their surface either solitarily or 2 or more together, but always separated by short, stem-like intervals, the uppermost being ciliate. The receptacles are subcylindric and ramified.

India : Tuticorin, Hare Island, in shallow water.

Geogr. Distrib. Admirality Isles, New Holland, Malayan Archipelago.

Sargassum Ag.

As I knew that Professor *W. A. Setchell*, Berkeley, California, was interested in the study of SARGASSUM from the East, I requested him to kindly determine for me my small collection of Indian SARGASSUM. He most kindly agreed to do so, and I have now received a list of his determinations. When he sent these back to me, he pointed out in his letter that his determinations must be regarded as only tentative until they are compared with the type specimens. As he was not

able to see the type specimens of these Indian species himself he asked me to compare my plants with the type specimens found in *J. Agardh's* Herbarium in Lund, Sweden.

In the list given below only 6 species are mentioned, of which four were gathered by myself and of which materials were sent to Professor *Setchell*, and two were found in a collection of Algæ belonging to the British Museum sent to me for determination. But many more species are known previously from India. In *J. Agardh's* Species Algarum, vol. I, Algas Fucoideas complectens, Lund, 1848, several species of Indian Sargassum are described; and in *R. K. Greville's* paper; "Algæ Orientales:—Descriptions of new species belonging to the genus Sargassum", in Annals and Magazines of Natural History, Ser. 2., vol. 3, p. 85, 14 species are mentioned as found in India.

Agardh's and *Greville's* works appeared almost at the same time, and, as Professor *Setchell* pointed out in his letter to me, here again is a question as to whether *Greville's* paper or *J. Agardh's* book was published first. "That would be a matter of the day and month of the year 1848, which may be difficult to settle", writes Professor *Setchell*. Fortunately we seem to be able to solve this question satisfactorily by means of the reviews of the two works. *J. Agardh's* work is reviewed in the Botanische Zeitung, 1848, p. 754, in the number published on October 27th, and must therefore have appeared before that date. On the other hand the first part of *Greville's* paper containing three species and Plate IV (the first of the 7 plates belonging to the paper) did not appear until 1849 according to "*Vickström's* Aarsberettelser" for 1849, p. 178. Accordingly *J. Agardh's* names have the priority ([1]).

Besides the two above mentioned papers dealing with SARGASSUM, I wish to mention also, as important works on this genus, *J. Agardh's*, "Species Sargassorum Australiæ descriptæ et dispositæ" (Kongl. Svenska Vetenskaps—Akad. Handl., Bd. 23, Stockholm 1889) and *A. Grunow's* most valuable posthumous work: "Additamenta ad cognitionem Sargassorum" in Verhandlungen d. k. k. zoologisch-botanischen Gesellschaft in Wien, 1916.

Sargassum tenerrimum J. Ag.

Agardh, J., Spec. Alg., vol. I, p. 305; Spec. Sargass. Austral., p. 83, Kützing, Spec. Alg., p. 626.

SARGASSUM CAMPBELLIANUM Greville, Sargassum, p. 88, Pl. V. *Grunow*, Additamenta, p. 370, no. 70.

---

[1] *J. Agardh's* work on the Phæophyceæ is already reviewed in the foregoing volume of *Vickström's* Aarsberettelser for the years 1845-48.

To this species (Fig. 6 and Plate II) I had referred already before I sent my collection to Professor *Setchell* several specimens gathered at Bombay from where, too, the type specimens in Herb *J. Agardh*, Lund, originate. I have been able to compare my specimens with *Agardh's* and I have stated their congruity. SARGASSUM CAMPBELLIANUM Grev. is surely closely related to and most probably identical with
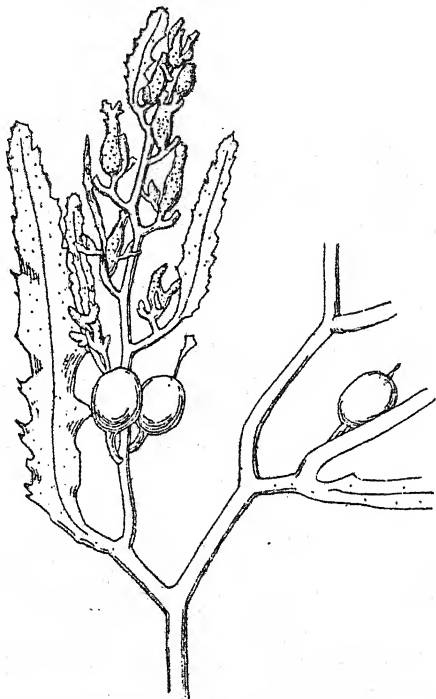

Fig. 6. **Sargassum tenerrimum** g. Ag. Part of the thallus with leaves, vesicles and receptacles. × 3.

S. TENERRIMUM J. Ag.[1]. In " Spec. Sargassorum " *Agardh* refers to S. CAMPBELLIANUM as a synonym of his species, and Prof. *Setchell* writes to me that the minor differences may be simply differences in stages of development. In *J. Agardh's* Herbarium I have not seen any specimens of *Greville's* plant, but several of my specimens seem to resemble his figures.

As the name indicates S. TENERRIMUM is a delicate, thin leaved, fine species which contrary to most species of this genus adheres to the paper when dried. The dried plant has a brownish olive-green colour.

---

[1] *Grunow*, l.c., refers to *Greville's* species as a var. CAMPBELLIANA (Grev.) of S. TENERRIMUM J. Ag.

The stem is glabrous and rarely more than ½ mm thick when dried. The leaves reach in the lower parts of the plant up to a length of 6 cm and a breadth of 1½ cm, but become gradually much smaller upwards. They are linear or linear-lanceolate in shape with a narrow rounded apex and an elongated cuneate base running evenly over
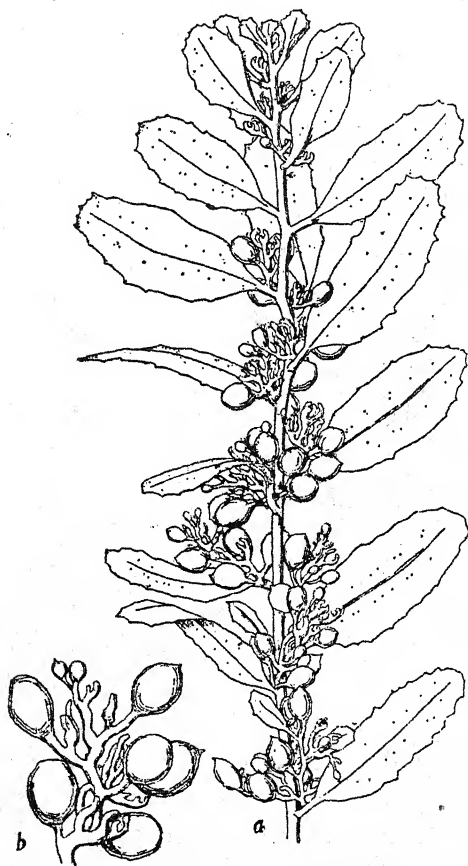

Fig. 7. **Sargassum cinereum** g. Ag. var. **berberifolia** Grun, a, part of the thallus with leaves, vesicles and receptacles; b, small part of the same more magnified a, × 1½; b, × 3.

into the short stipe. Their margin is sinuate-dentate. The leaf is thin, translucent, with a thin midrib and on both sides scattered small cryptostomes. The vesicles are nearly spherical, about 3–5 mm long and about just as broad, their stipe being about half as long. The receptacles are androgynous, ramified and spinose.

India : Bombay, in many localities in the open sea ; Dwarka.

Geogr. Distrib. Arabian Sea along the Indian Coast.

Sargassum cinereum J. Ag. var. berberifolia Grun.

Grunow, A., Additamenta, p. 378, no. 76.

The determination of this plant (Fig. 7 and Plate III) I owe to Professor *Setchell*. As no specimens of this variety are to be found in Herb. *J. Agardh*, in Lund, I have only been able to compare my plant with specimens of forma *typica* of which several specimens from Hongkong are found in *J. Agardh's* Herbarium. These have smaller leaves than mine, whereas the vesicles have nearly the same size. Professor *Setchell* writes to me that he has tentatively referred this plant to SARGASSUM CARPOPHYLLUM, but he adds that the leaves are too broad for that species, at least according to the description. Therefore I looked up this species in Herb. *J. Agardh*, Lund, and found that it had much smaller leaves and was on the whole rather different from my plant.

The specimens in my collection are of a greyish-brown colour. The stipe is glabrous. The leaves are about $2\frac{1}{2}$ cm long and $\frac{3}{4}$ cm broad in the fruiting part; in a young plant the leaves are about $3\frac{1}{2}$ cm long and 8 mm broad. The leaves have an obtuse apex and a cuneate base passing over into a short stipe; their margin is rather distantly sinuated or toothed; scattered cryptostomes are found on both sides of the midrib. The vesicles are nearly spherical or somewhat longer than broad, up to about $\frac{1}{2}$ cm long and 3–4 mm broad, but many are smaller. The receptacles, male and female on different plants, are small branched panicles without spines.

India : Karwar in tranquil bays.

Geogr. Distrib. Indian Ocean.

Sargassum Wightii (Grev. mscr.) J. Ag.

Agardh, J., Spec. Alg., vol. I, p. 329 ; Species Sargass. Austral., p. 86. *Greville, R. K.*, Sargassum, p. 95, tab. X. *Grunow, A.*, Additamenta, p. 382, no. 81.

In a collection of Algæ belonging to the Herbarium of the British Museum some specimens are to be found which seem referable to this species. The specimens are sterile or just beginning to fruit. There is a narrow-leaved and a more broad-leaved form present. The narrow-leaved plant is when dried almost black with thick opaque leaves. These are nearly linear, up to about 5–6 cm long and 2–3 mm broad tapering towards both ends, the apex is acute or obtuse, at the base attenuated, passing evenly over into the short stipe 1–3 mm long. The margin of the leaves is nearly entire or a little sinuate. The midrib is scarcely seen and there are very few cryptostomes on both sides of it. The vesicles are rather large, oblong up to about 7 mm long and 4 mm broad and have a rather long stipe up to

about 7 mm long; it is seldom that they end in a long tip. The receptacles are, as mentioned above, only just beginning to be developed. This specimen most probably belongs to forma SUBLINEARIS Grun., which is known from Ceylon.

The other specimen has broader, thinner and much more brown leaves about 5-6 cm long and 8-9 mm broad; the margin of the leaves is sinuate-dentate. The vesicles are nearly spherical and much smaller, about 5 mm broad. The specimen is sterile. This plant has some likeness to a specimen from Ceylon (*Harvey*, Ceylon Algæ no. 106) found here in the Botanical Museum, but the colour of the Indian plant is much more brown and the vesicles smaller than those of the Ceylon specimen.

India: Pamban, *M. O. P. Iyengar*; without locality, *Wight*.
Geogr. Distrib. India, Ceylon.

Sargassum ilicifolium (Turn.) C. Ag. var. venusta Grun.?

Grunow, Additamenta, p. 404, no. 106.

Professor *Setchell* has put a? after the name and says about the determination : " A specimen which agrees fairly well, but not exactly, with a specimen in our Herbarium determined by *Grunow* and sent out from his collection. The coincidence is not exact, but I presume that these Indian forms are mostly of this variety ".

The leaves in the specimens (Plate IV) are about 3½ cm long and 1 cm broad decreasing upwards; the margin of the leaves is sinuate-dentate. Several scattered cryptostomes are found on both sides of the midrib. The vesicles are spherical up to about 3 mm broad, most of them being smaller. The receptacles are fastigiately branched, triangular and spinulose. According to *Greville* they are androgynous, but I found only female ones.

India : Karwar. Bengi Bay in a rather exposed locality. At Bombay (Bandra seaface) I gathered a single young female specimen coming closer to the typical form with sharply serrate or dentate leaves.

Geogr. Distrib. Indian Ocean, Red Sea.

Sargassum myriocystum J. Ag.

Agardh, J., Spec. Alg., vol. I, p. 314; Species Sargass. Austral., p. 99.

Grunow, A., Additamenta, p. 440, no. 134.

To this species, which according to *J. Agardh* is already known from India, I think a female specimen from the collection of the British Museum can most probably be referred, as it seems to agree rather well with *Grunow's* description.

The stem of the specimen is rather rough with short processes. The leaves are about 2 cm long and ½ cm broad, decreasing upwards and becoming quite small higher up. They are dark, nearly blackish-brown when dried. Their margin is more or less dentate, and their apex more or less rounded.

Scattered cryptostomes are present on both sides of the midrib. The vesicles are small, spherical, 1–2 mm broad. The female receptacles are somewhat spinulose and very much ramified.

India : Pamban, *M. O. P. Iyengar :* without locality, *Wight.*

Geogr. Distrib. China, Java, India.

Sargassum plagiophyllum (Mert.) J. Ag.

Agardh, J., Spec. Alg., vol. I, p. 309 ; Species Sargass. Austral., p. 120, Tab. XII. *Grunow, A.,* Additamenta, p. 9, no. 149.

I owe the determination to Professor *Setchell.* I have compared my specimens with a specimen from "Detroit de Malacca " (Baume) in Herb. *J. Agardh,* Lund. This is rather a poor specimen, being only the uppermost part of a plant, with somewhat smaller leaves than those in my plant. My specimens (Plate V) on the other hand agree quite well with *J. Agardh's* figure.

The stipe in my specimens is terete, glabrous, about 1 mm thick (dried). The leaves are linear-oblong, dull, with subundulate or subdentate, often almost quite entire margins with obtuse apex and acute base passing evenly over into the quite short stipe. In the lower parts of the specimens the leaves are about 3 cm long and ¾ cm broad becoming smaller upwards and have scattered cryptosomes on both sides of the midrib. The vesicles are small, spherical, about 2 mm broad, sometimes tipped. The only fruiting specimen is a male plant; the branched receptacles have no spines, whereas the female ones according to *Grunow's* description are provided with spines.

India : Karwar in tranquil bays.

Geogr. Distrib. Indian Ocean.

Turbinaria conoides Kütz.

Kützing, Tab. Phycol., vol. X, p. 24, tab. 66, II e, f. *Barton, E. S.,* A systematic and structural account of the genus Turbinaria, Lamx. in Transact. of the Linnean Society of London, 2nd Ser., Botany, vol. III, part 5, London 1891.

TURBINARIA VULGARIS, var. CONOIDES J. Ag., Spec. Alg., vol. I, p. 267.

In her above-mentioned monograph of the genus TURBINARIA Miss *Barton* quotes as synonym to this species FUCUS TURBINATUS L., (Spec. Plant., vol. II, p. 1160), and bases this statement upon a

specimen found in *Linné's* Herbarium, London.   Consequently in my paper on the *Forsskaal* Algæ [1] I pointed out that *Kützing's* specific name for this species ought to be changed into *Linné's*, namely, TURBINARIA TURBINATA (L.).   But I later on found that for several reasons this cannot be done, first because *Kuntze*[2] has already used this name for the West Indian species, TURBINARIA TRIALATA, and secondly, because, as Mr. Geoffrey Tandy, British Museum, London, who is much interested in the question, has kindly informed me, there was no specimen of TURBINARIA in Herb. *Linné* before 1767, *Linné's* FUCUS TURBINATUS in "Spec. Plant" (1753) being based only upon the West Indian plant described in *Hans Sloane's* "Natural History of Jamaica," 1707, p. 58, tab. 20, fig. 6.

Accordingly the name of this species must remain as proposed by *Kützing*.

Of this plant we have here in the Botanical Museum an old Indian specimen (var. TYPICA, with vesicles) originating from the former Danish colony, Tranquebar, in South India.   Moreover, in a collection of Indian Algæ belonging to the British Museum, London, and sent to me for determination a specimen from Pamban is present ; it belongs to var. EVISICULOSA Barton, as the stipe of the leaf is not inflated.

India : Tranquebar, *J. P. Rottler* ; Pambam, *M. O. P. Iyengar*.

George Distrib.   Sumatra, Singapore, Ceylon, Java, Celebes, China, Australia.
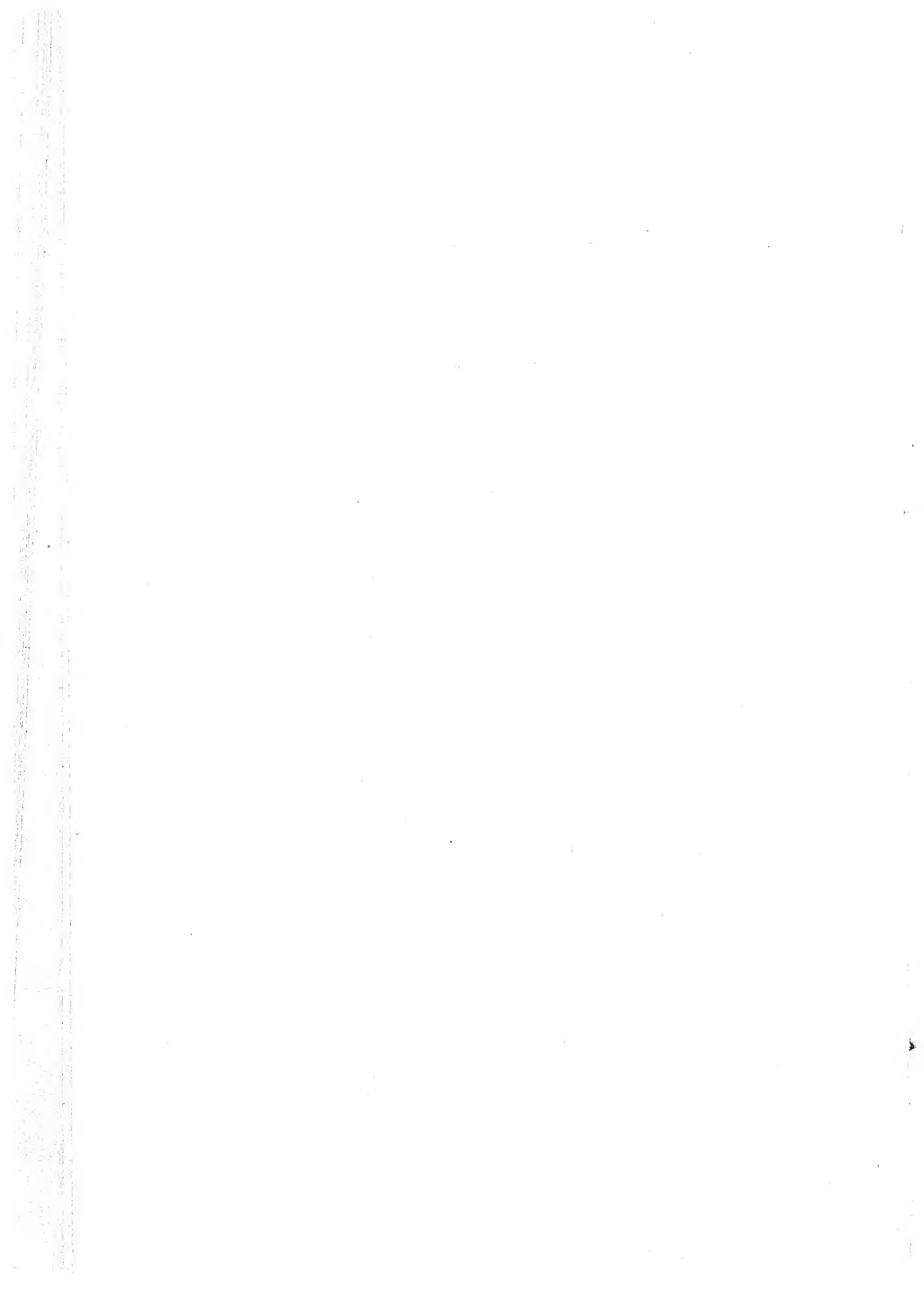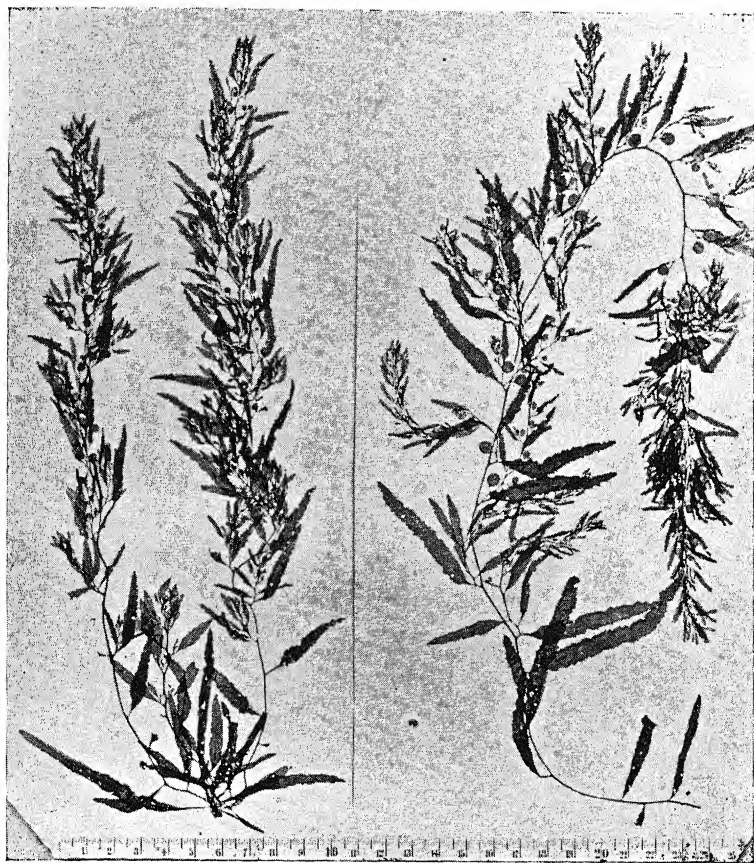
COPENHAGEN,
June, 1932.

---

[1] *Boergesen, F., A* revision of *Forsskaal's* Algæ mentioned in Flora Aegyptiaco-arabica and found in his herbarium in the Botanical Museum of the University of Copenhagen (Dansk Bot. Arkiv, vol. 8, 1932, no. 2, p. 12, the note)

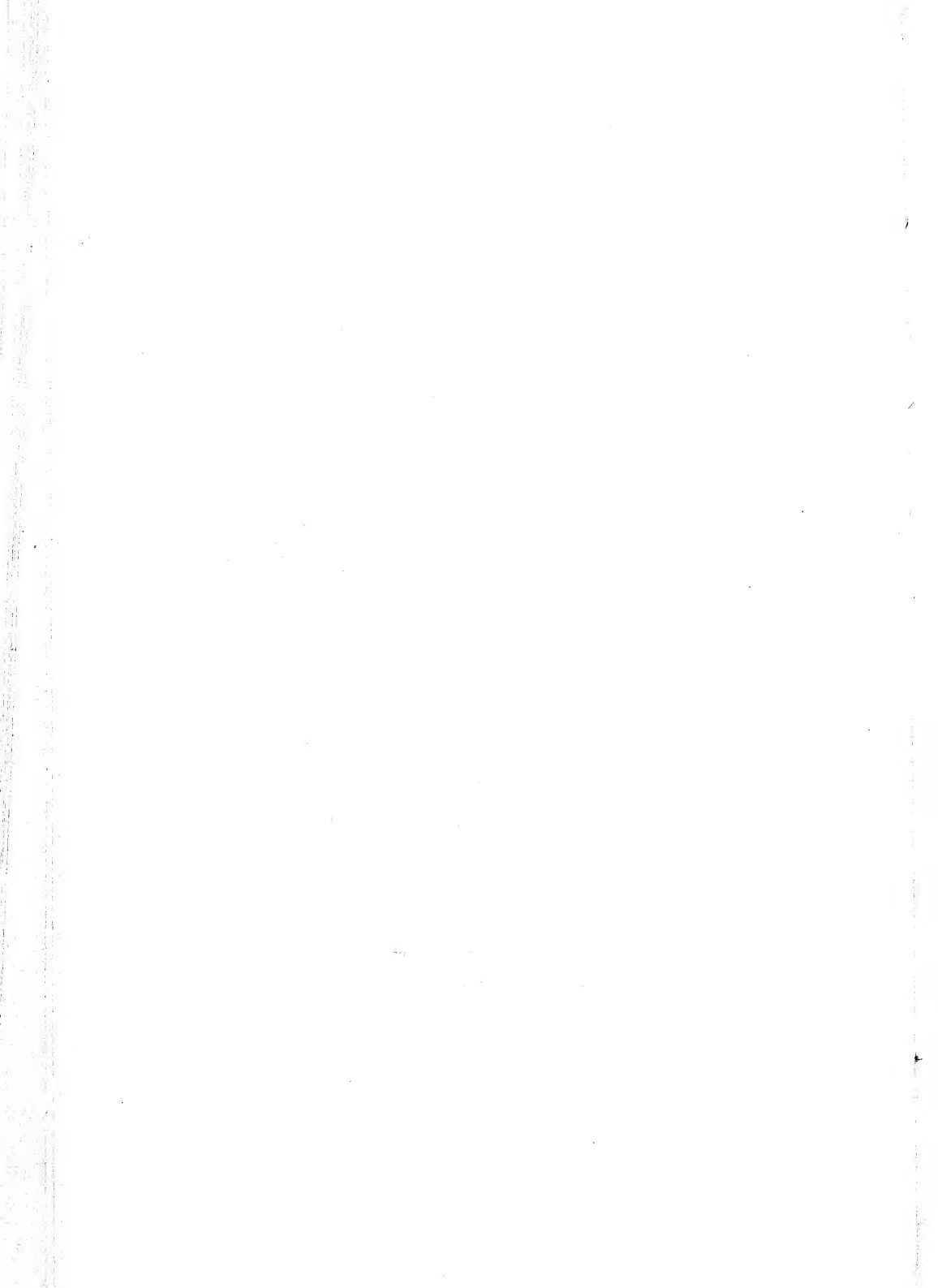[2] *Kuntze, O.,* Revisio generum plantarum, Pars II, p. 434.

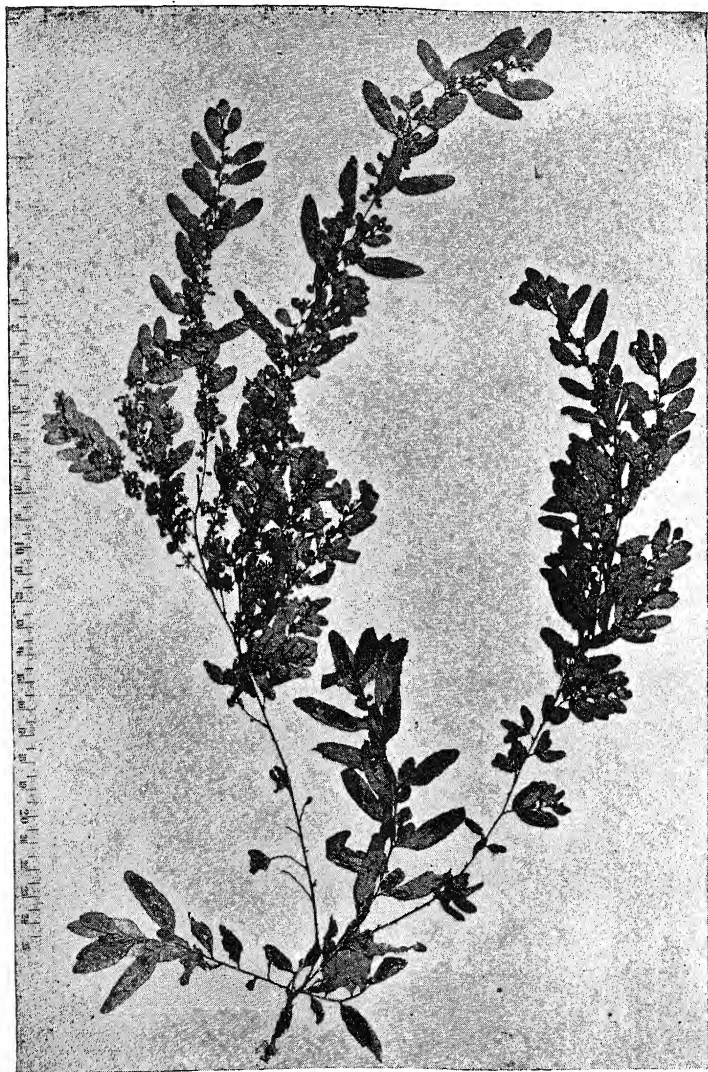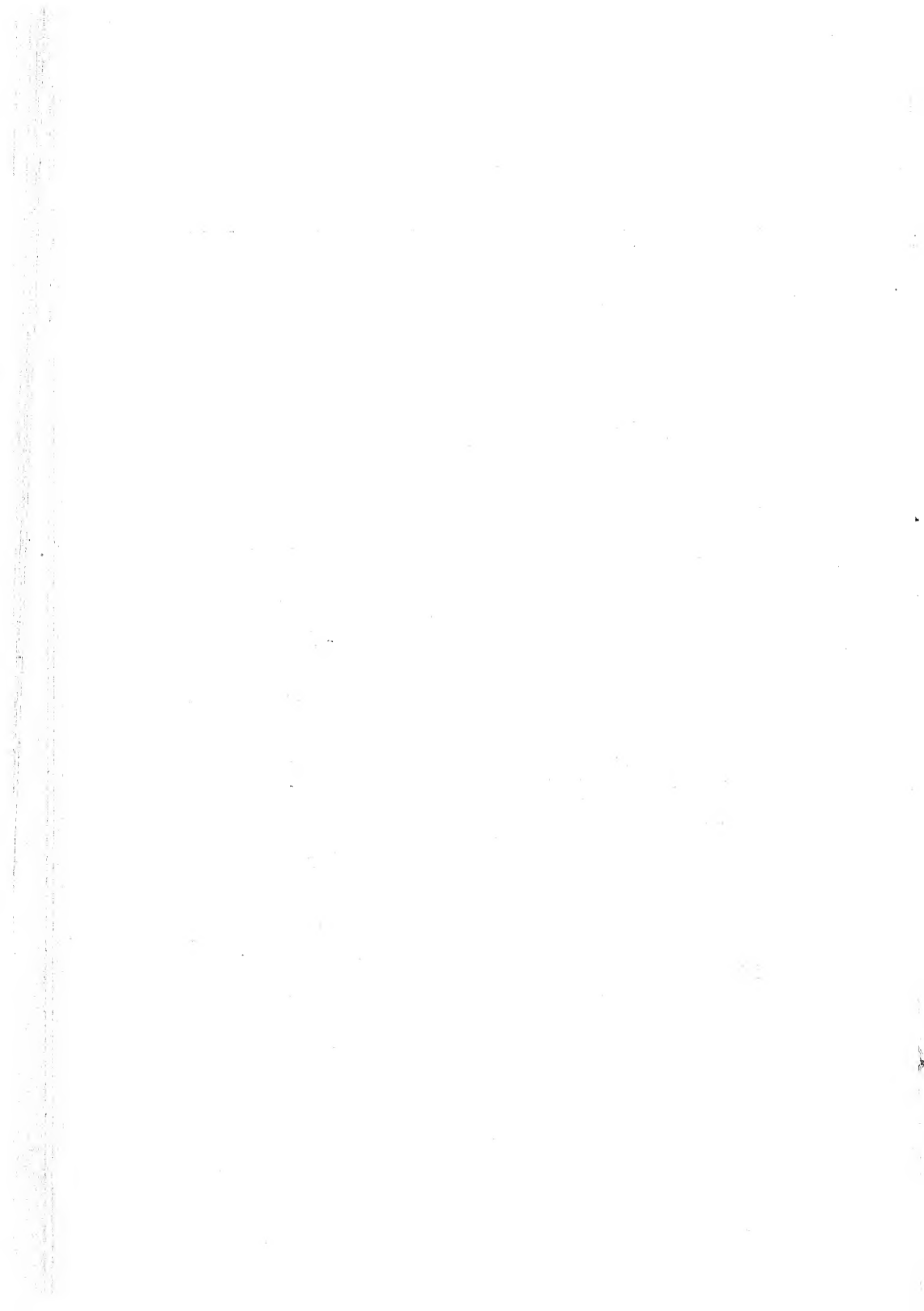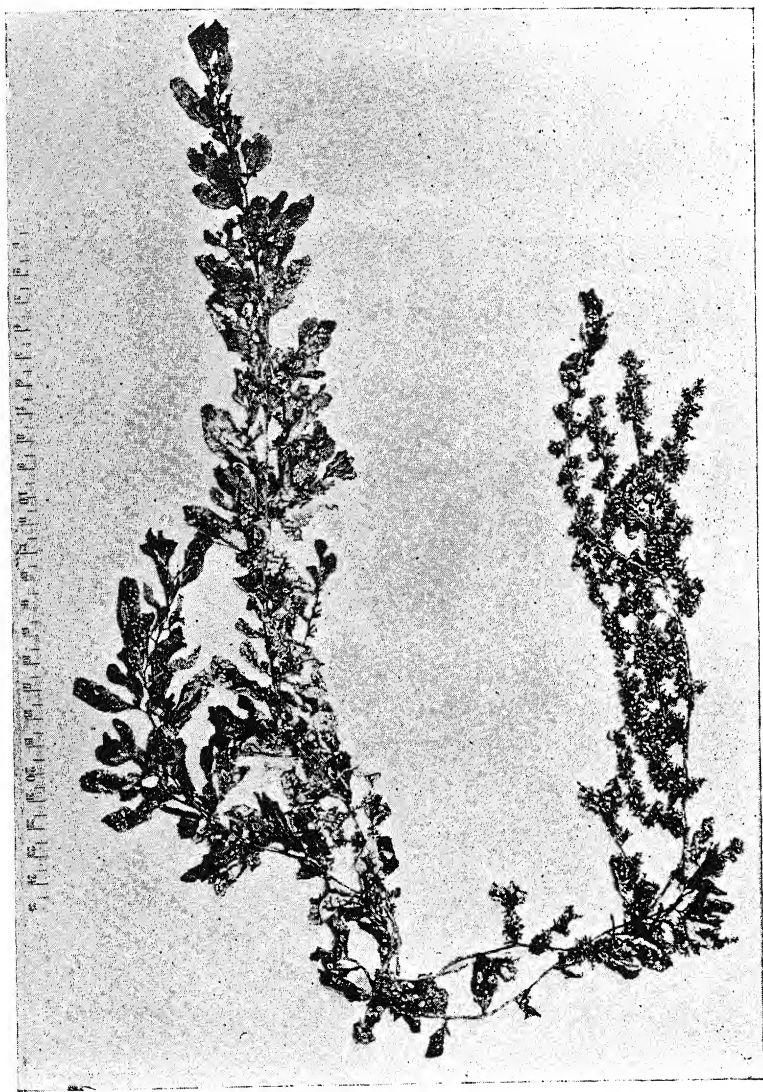**Struvea tuticorinensis** Boergs. sp. nov. A tuft of the plant. × 3.
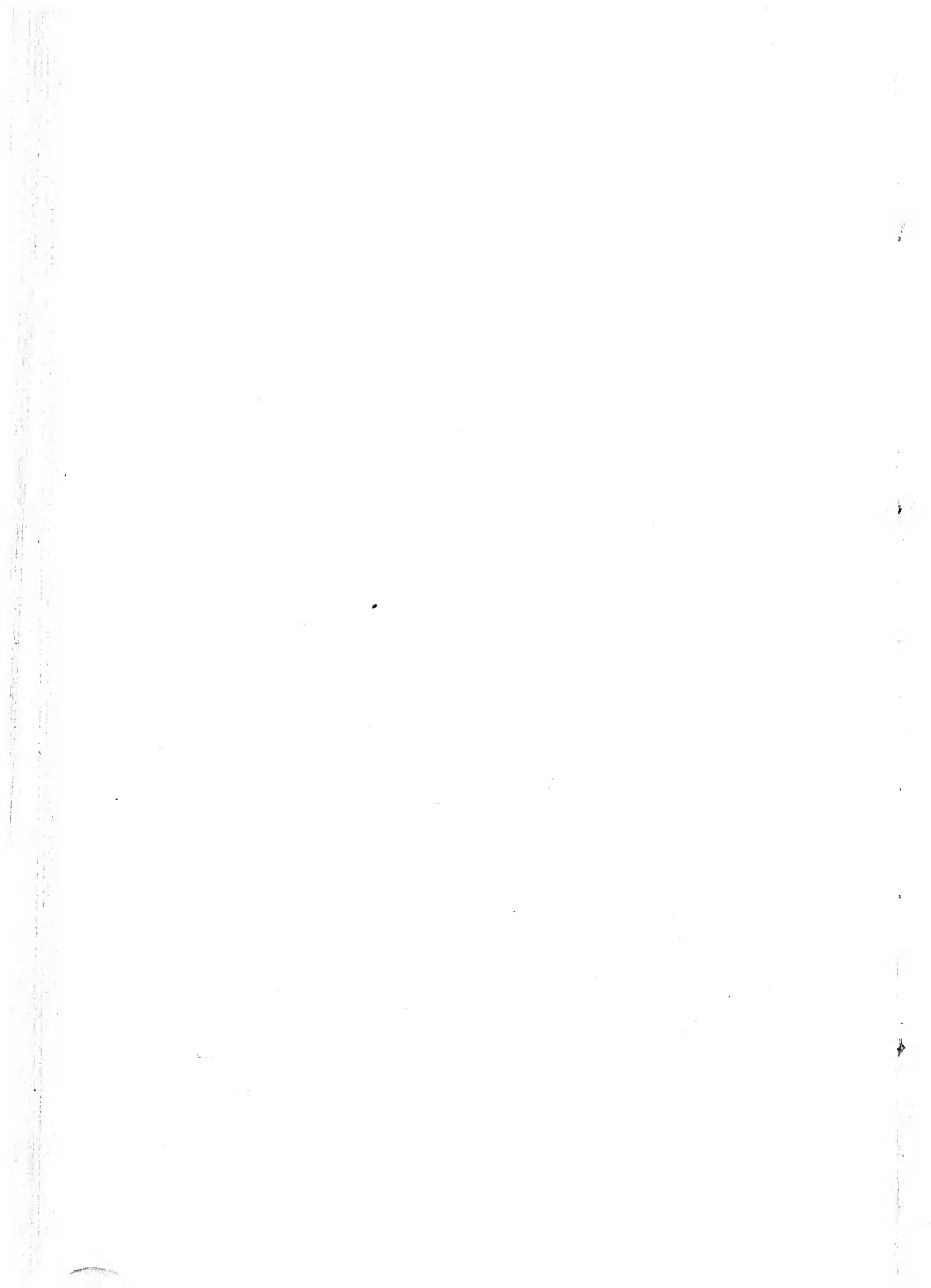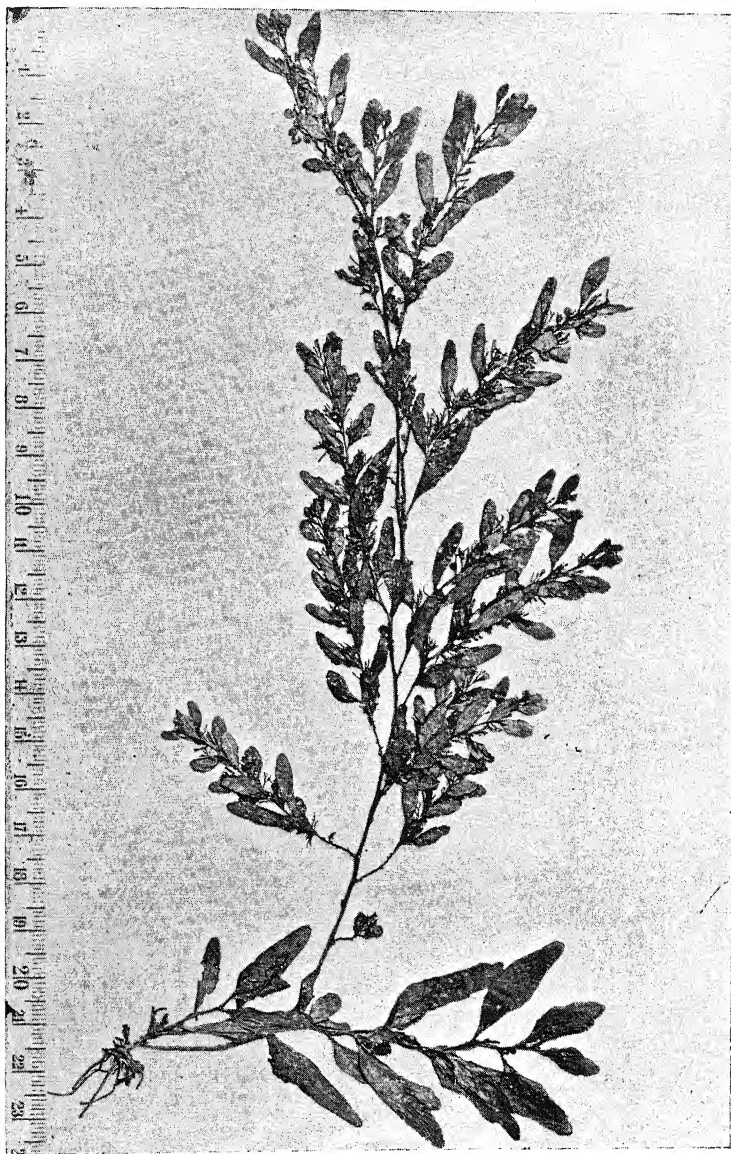
Sargassum tenerrimum J. Ag.
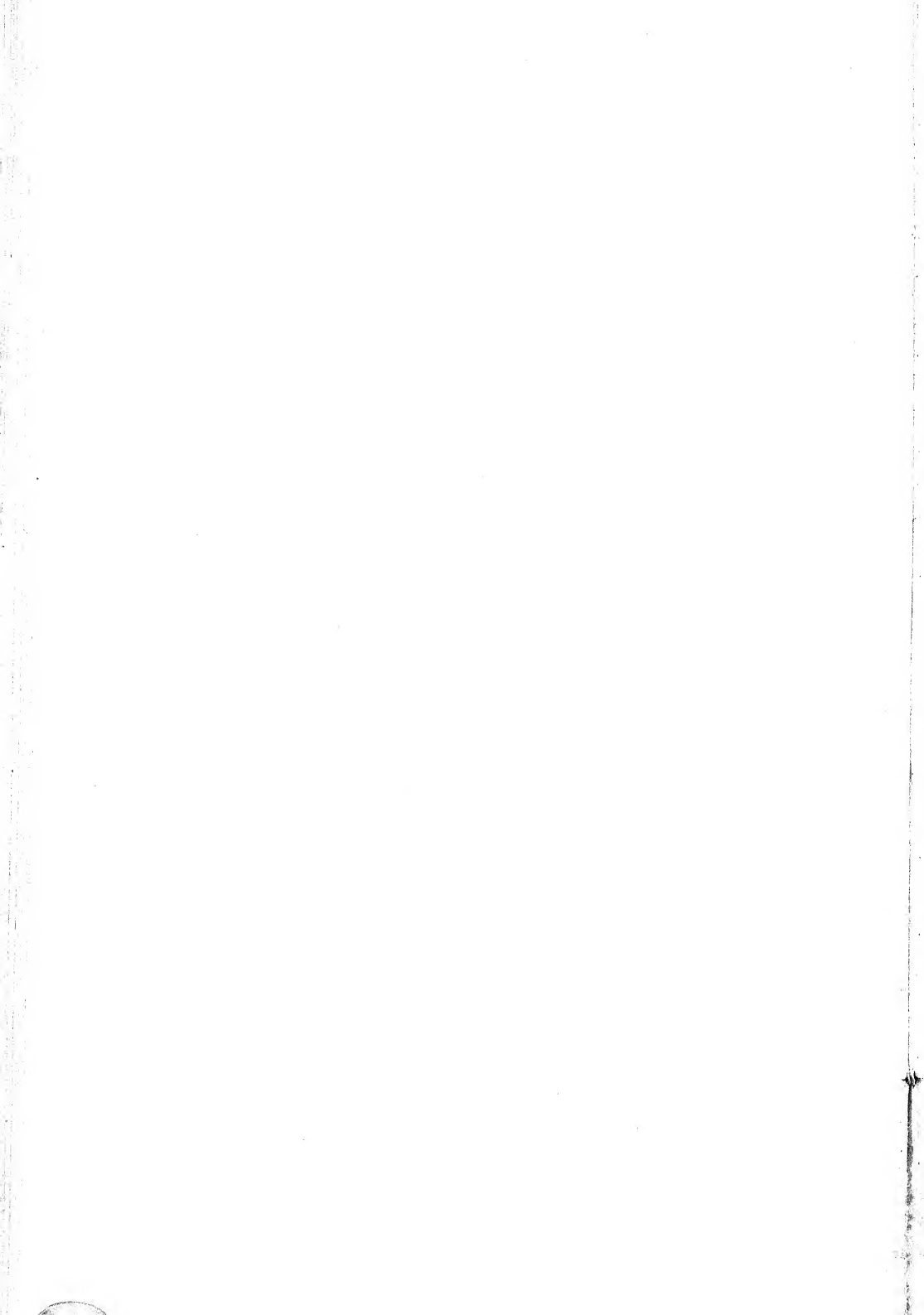
Sargassum cinereum J. Ag. var. berberifolia Grun.

Sargassum ilicifolium (Turn.) C. Ag. var. **venusta** Grun. ?

Sargassum plagiophyllum (Mert.) J. Ag.

CHAROPHYTE NOTES FROM AGRA, U. P.

BY

G. O. ALLEN, I.C.S.

It is hardly necessary to describe the geographical situation of Agra as it must be known the world over from its fascinating Taj Mahal. In a scientific botanical journal however it should be mentioned that its latitude is c. 27° N and longitude c. 78° E and also that its average rainfall is 26 inches. It lies in the S.-W. corner of the U. P. and in the opinion of the writer of the local Gazetteer it may probably claim to be the warmest station in the U. P. After nearly two years' experience of the district I feel it is almost like living on the edge of a rocky desert, with one redeeming feature in the mass of delicate blossom of that widespread xerophyte, the karil (*capparis aphylla*). The dry climate, low rainfall and the excellence of the natural drainage afforded by the Jumna and its tributaries and by the Chambal have resulted in a great scarcity of lakes and marshes. Even the ponds are of such a temporary nature that they are mostly dried up before the cold weather sets in. During the rains though I visited a number of pools I found them all very muddy and did not succeed in finding a single species during this usually profitable season of the year. Except for the Chambal and the Jumna, the only permanent piece of water I came across was Kitham reservoir eleven miles along the Muttra-Delhi road.

Agra therefore afforded a complete contrast to my other two hunting-grounds, Gonda and Saharanpur: in fact such interest as my notes may have is rather of a negative kind, a record of what I did *not* find. Instead of 22 species as at Saharanpur my total for two cold weathers and one "rains" amounted to no more than 5, 1 Nitella and 4 species of Chara. One of these latter, however, proved to be an addition to the Indian list. They are as follows : *N. hyalina, C. aspera, C. fragilis, C. contraria* and *C. vulgaris*.

I had not much occasion to visit the Jumna. Its banks were sandy and the only species I found along its edge was *C. contraria*.

Kitham reservoir is a fine expanse of water but disappointing from my point of view. Being of too permanent a character there was little variety in the nature of the shore and the depth round the edge remained fairly constant. All except *C. aspera* were to be found here. The only point of interest I noticed here was in connection with

C. vulgaris. I first found it in a little stream that issued from the base of a high embankment. In some small pools it was so very ecorticate that I thought I had run across something new to me until I realized later that it was only the cramped conditions that were responsible for this as in deeper water it appeared in a normal state.

Mr. Groves tells me that Mr. T. B. Blow, that indefatigable searcher after charophytes, once collected in the Taj gardens a Chara that appeared to be *C. vulgaris.* I could find no trace of it however in that pretty little marble tank from the platform of which so many thousands have gazed on the wondrous tomb.

For the Chambal with its clear blue water I have a great affection. For one thing it was here that I found what is probably *C. aspera* but that is not the only reason. After a road journey of 35 miles it was very pleasing to pass down a deep cleft and find that peaceful expanse of river with its picturesque pontoon bridge suddenly open out before one.

The river runs considerably below the level of the surrounding country (as is usual in this rocky part of India) and hence arises that wonderful system of ravines as much as a mile broad in some places. Incidentally they afford a capital hiding-ground for dacoits as it must be only after long acquaintance with it that one can readily find one's way about such a maze.

This deep-cut bed also afforded a novel and pleasing sight for a dweller in the Gangetic Plain : for here was exposed a section showing a succession of four different strata, the lowest of which furnishes road metal that was being quarried busily at the time. These strata I take to be, in order from the top, the Vindhyan (Kaimur) Sandstone Kaimur Conglomerate, Gwalior Sandstone and Bundelkhand Gneiss. This charming spot is reached by crossing Dholpur State along the road to Bombay and it was in a deep pool amongst the rocks on the left bank that I found my Chara, perhaps the first record for this group from this State.

C. aspera is a slender plant generally found growing along with *C. fragilis* to which though an altogether smaller plant it bears outwardly a good deal of resemblance. In British Charophyta[1] it is referred to as being of a less intense green than *C. fragilis.* I found the upper part of the stem quite orange in hue and with its quantity of bright red antheridia the general effect of the plant was very pretty. The oogonia are also orange at first before turning black as the oospores ripen. The plant was much lime-incrusted and decidedly

---

[1] Groves and Bullock-Webster : Ray Society.
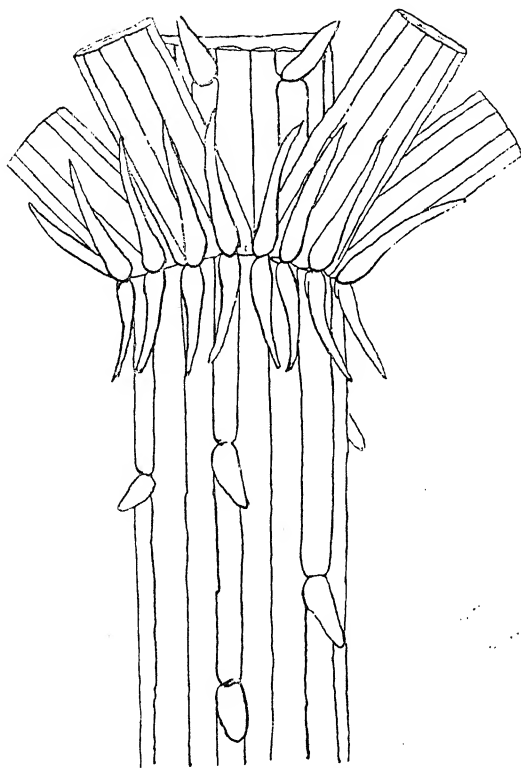
C. aspera   × c. 125

brittle. *C. fragilis* is also one of its nearest relations in India as they both belong to the small group of triplostichous species of Chara though *C. aspera* is readily distinguished by its diœcious character. There is one other diœcious triplostichous plant recorded from India viz. *C. infirma* though in Notes on Indian Charophyta[1] it is spoken of as rather a doubtful species. Its rudimentary spine-cells serve to distinguish it from *C. aspera*.

C. aspera has been recorded from Central Asia (Turkestan) but not from India before. The species is fairly common in England and Europe generally. As Mr. Groves points out the Indian plant differs from the English in having small roundish spine-cells instead of long spines and in the cortex being more regularly triplostichous.

A very characteristic feature of this species is the production at the lower nodes of almost spherical whitish bulbils which, as remarked in British Charophyta, no doubt account for its not fruiting very freely. Incidentally that work also speaks of gametangia at the lowest 2–3 nodes : in my plant antheridia are produced at the fourth branchlet-node as well. The stipulodes are well developed (not unlike those depicted in fig. 3, plate XXXIX British Charophyta) : hence it is somewhat curious to find the spine-cells so small.

The first year I found this plant on 3rd February but as I had no chance to gather more of it I kept a good look-out the following year. I found no signs of it till 18th January when it was yet in a very young state. Both years I gathered it in exactly the same pool, which being deep and rocky presumably admitted of the oospores remaining undisturbed by the heavy currents in the monsoon. I continued to find it the second season till the end of March. Along the banks of the Chambal were also growing *N. hyalina* and *C. contraria*.

---

[1] Journal of the Linnean Society (Botany) Vol. xlvi, April 1924.

PERENNATION AND VEGETATIVE REPRODUCTION IN *ZEUXINE SULCATA* LINDLEY

BY

AMAR CHAND JOSHI,

Department of Botany, Benares Hindu University, Benares.

(With 1 figure in the text.)

———

Zeuxine sulcata Lindley is a very widely distributed orchid in Asia. It is found in Afghanistan, India, Java, China and Philippines. Hooker (1) describes it as the commonest Indian orchid, it being met with throughout the plains and the low hills of the country from Punjab and Sindh to Assam and Chittagong and southward to Ceylon.

In the plains of northern India this orchid grows during winter. Then green aerial shoots with erect grass-like membranous leaves are produced. These mostly range between 2 to 6 inches in height. The erect stem has sometimes a creeping base and there it bears a number of fleshy roots. The shoots ultimately produce terminal spikes of small white flowers. From these are produced capsules containing a very large number of seeds. These ripen and are shed towards the end of winter and after this these aerial shoots also wither and die away. The orchid passes the summer underground and new green aerial shoots are again produced on the advent of the next winter season. Nothing, however, is known about the condition in which the orchid passes the unfavourable season, even though it is so common all over the country.

Last year during the month of April, the writer was on a short holiday to a place called Hoshiarpur in the sub-himalayan tracts of Punjab. Here plants of this orchid were found growing in a small grassy plot on the bank of a rainy season stream, which is locally known as 'Cho'. The place was at a distance of about three miles from the town. It was just the beginning of summer. The orchids had already fruited and the seeds had been shed and they were in various stages of withering. As the time appeared to be opportune, it was decided to look these up for their mode of perennation. For this purpose a few plants were dug out from the ground along with the soil and brought home. There these were placed in a bucket of water and the soil was gradually and carefully removed, so that there was no danger of any parts getting broken. One such plant

with complete underground parts is sketched in the accompanying text-figure,—the aerial portion had mostly died away ; and it clearly reveals the mode of perennation of the orchid.

It appears that in this orchid towards the close of the growing season, and before the advent of the hot dry season, certain underground branches are formed from the basal nodes of the main stem. These are marked Br1, Br2, and Br3 in the figure. They are of the
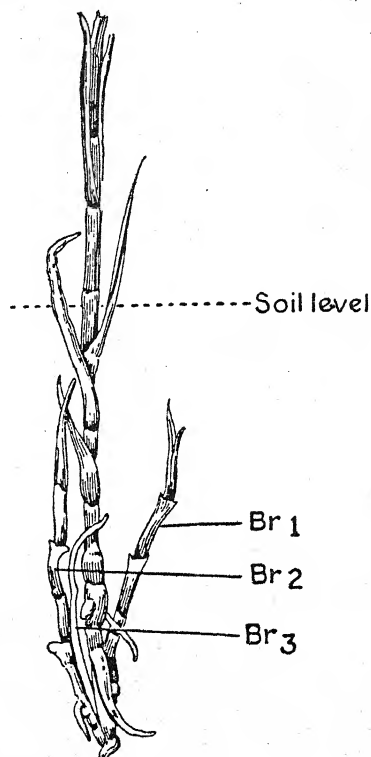
Zeuxine sulcata. A plant dug out from the soil during the month of April The parts below the line marked soil level are all underground. Br1, Br2 and Br3 are the perennating underground branches. *Normal size*

ordinary form and become fairly large, often producing 6 or 7 internodes and short membranous scaly leaves from the nodes. Often they develop their own adventitious roots. In some cases a root is formed on the node of the old axis of the plant just below such a branch. In colour, these new branches are nearly white, while the main stem is more or less brownish. Sections of the various parts of the plant showed that all the food material is transferred to these new branches and their cortex becomes packed with solid carbohydrates.

After these branches have been formed, the main stem withers and dies away. Only these branches packed with food material are left behind in the soil. They never come to the surface and act as the means of tiding over the unfavourable season. On the advent of the next growing season, however, they are probably again stimulated to activity and grow up into new plants.

The form and disposition of these underground perennating branches suggests why these have been overlooked up to this time. The plants of *Zeuxine sulcata* usually grow in fairly compact clay soil. In such a soil if the main flowering shoot is pulled out, the underground branches will be always, on account of their position in the soil, left behind. Since this is the usual method of taking out of small herbaceous plants, this may account for the non-observance of these structures so far.

The number of such perennating branches on one plant may be one, two or more. In the specimen figured, there are three such branches. As each of these can grow up into a new plant, more than one offspring may be produced from one individual. So these underground branches also serve as a means of vegetative reproduction and multiplication. Many times in nature plants of *Zeuxine* are found in small clusters. Their method of vegetative reproduction explains this.

Discussion.

The method of perennation of *Zeuxine sulcata* is of great interest on account of its extreme simplicity. In no other terrestrial orchid, a method simpler than this has been recorded. In the majority of such orchids, only buds are laid toward the end of the growing season and they develop into shoots only on the advent of the next season. Here, however, they begin their development in the same season and form fairly big underground branches. The common feature between the two cases is that the green aerial shoots develop only at the beginning of the next favourable season and the unfavourable season is passed entirely underground. In the majority of terrestrial orchids during this period food is stored either in internodal or root tubers, but in *Zeuxine sulcata*, it is directly transferred to these underground branches which have to grow into aerial shoots later on and there are no such special organs for this purpose. The form of these underground branches is also just ordinary and shows no special peculiarities.

The mode of perennation of *Zeuxine sulcata* is of further interest on account of the possible light that it may throw on the origin of more complex methods of perennation found in the terrestrial orchids,

especially on the origin of such complex tubers as those of Ophrydeae (*Ophrys, Orchis*, etc.). According to Rendle (2) the tubers of this tribe "consist of next year's stem-bud, which has united very early with the fleshy adventitious root standing exactly beneath it." It has been mentioned above that sometimes in *Zeuxine*, there stands an adventitious root on the same node from which a perennating branch arises and exactly beneath it. It may thus represent the simpler condition from which the more complex tubers of Ophrydeae may have been derived by postponement of the development of the branches for sometime, tuberisation of the adventitious root and the fusion of the two.

Summary.

The method of perennation of *Zeuxine sulcata* is very simple. Towards the end of growing season, certain underground branches are formed from the basal nodes of the main stem and food material is stored up in these. They act as the perennating organs of the plant during the unfavourable season and grow up into green aerial shoots on the advent of the next growing season.

BENARES,
September 8th, 1932.

Literature Cited.

1. HOOKER, J. D.—The Flora of British India. Vol. V. London, 1890.
2. RENDLE, A. B.—The Classification of Flowering Plants. Vol. I. Cambridge, 1930.

THE DISTRIBUTION OF WILD CONIFERS IN THE INDIAN EMPIRE

BY

KALIPADA BISWAS, M.A.,

(Offg.) Superintendent, Royal Botanic Garden, Calcutta.

The study of the 'Distribution of Wild Conifers' in the Indian Empire has been undertaken at the suggestion of Dr. Birbal Sahni, Professor of Botany, Lucknow University, to whom I offer my grateful thanks. The present paper on living Conifers forms more or less a counterpart of Dr. Sahni's account of the Fossil Conifers in this country as described in his paper on the "Revision of Indian Fossil Plants" Part I "Coniferales" (Memoirs—Geological Survey of India Palœontologia Indica—New Series—Vol. XI, 1928). It is expected that these two papers may throw some light on the problem of the early history of the Indian Conifers and their distribution in the Indian Empire.

The distribution of wild conifers in the Indian Empire is mostly confined to the Western and the Eastern Himalayas including the hills of Northern Burma. The number of species is not very large but the Himalayan Conifers are well represented both in the Western and the Eastern Ranges, although predominating in the North-Western Mountains. J. D. Hooker in his Flora of British India enumerates thirteen genera and twenty-five species of which twenty-three species are considered wild. Of these twenty-three species, *Agathis loranthifolia* Salisb, and *Dacrydium elatum* Wall., are natives of the Malay Peninsula.

The following list of the species together with the localities, names of original collectors and dates of collections as far as available, has been compiled from the notes on the sheets of the Herbarium specimens of Conifers, collected from the different parts of India, Burma and Ceylon. These species have been described by J. D. Hooker under Conifereae in the Flora of British India, Vol. V, pp. 643–653 (1890). The species considered wild in this paper are those which are mentioned as such by Hooker and other earlier writers, and not marked "Cultivated" on the sheets of the Herbarium of the Royal Botanic Garden, Calcutta. The accompanying map (Plate I) illustrates the

range of distribution of the wild conifers in India. Some of the common wild conifers growing in different parts of the Indian Empire have been graphically represented thus :—

EAST HIMALAYA.   WEST HIMALAYA.

EAST HIMALAYA:

JUNIPERUS PSEUDOSABINA.

JUNIPERUS RECURVA.

LARIX GRIFFITHII.
ABIES WEBBIANA.
PICEA MORINDA.
TAXUS BACCATA.
TSUGA BRUNONIANA.

CEPHALOTAXUS GRIFFITHII.

CEPHALOTAXUS MANNII.
PINUS KHASYA.
PINUS LONGIFOLIA.

PINUS MERKUSII.

PODOCARPUS NERIIFOLIA.
PODOCARPUS LATIFOLIA.

WEST HIMALAYA:

JUNIPERUS PSEUDOSABINA.
JUNIPERUS RECURVA.
JUNIPERUS MACROPODA.
JUNIPERUS COMMUNIS.
CUPRESSUS TORULOSA
ABIES WEBBIANA.
PICEA MORINDA.
PINUS EXCELSA.
TAXUS BACCATA
TSUGA BRUNONIANA.
PINUS GERARDIANA.
CEDRUS LIBANI
VAR. DEODARA.

PINUS LONGIFOLIA.

General Distribution.

Although present state of our knowledge of the Floras of Nepal, Bhootan, Burma and Chino-Tibetan frontiers in the East and Northern Persia, Afghanistan and Baluchistan in the west does not allow us to make a detailed survey of the distribution of conifers in and about the Himalayas, I think it would not be out of place to discuss here the general trend of distribution of Indian conifers as far as the Herbarium material and literature permit. The range of distribution of the wild conifers as far as the records of the Herbarium sheets show tallies

mainly with the notes available in the literature.   From the data at my disposal one is led to draw the following conclusions regarding the occurrence of the individual species of the Indian conifers in their natural habitats. *Cupressus torulosa* appears to be the only wild Indian cypress mainly confined to the outer ranges of the North-Western Himalaya extending up to Central Nepal (as reported by Don from Webb's collection) and perhaps slightly beyond in its original wild state of growth ascending from [1] (5,000) to 13,600 feet.   Its occurrence is distinctly noticed between 6,000 to 9,000 feet.   The presence of this species in the Western Himalaya and then in W. Szechuan, China where it is said to be common, suggests its extension beyond Nepal across Bhootan to the Chino-Tibetan borders, although, Hooker limits, its extension up to Nepal.   The gap between Nepal and the Western boundary of Szechuan appears to be due to our insufficient knowledge of the flora of this important part of the land.   It ascends from 5,000 to (14,000 feet), being abundant between 10,000 and 13,000 feet.   *Juniperus communis* spreads mostly over the North-Western Himalayan ranges from Afghanistan to Kumaon from 5,500 to 14,000 feet in altitude. Kumaon may be considered the eastern limit of its wide range of distribution from Northern and Central Europe and the Coastal regions of the Mediterranean Sea, through Asia Minor to the North-Western Himalaya.   Its occurrence has also been noted in the United States and Canada.   *Juniperus pseudo-sabina* and *Juniperus recurva* are purely natives of the Himalayas occurring in higher altitudes. ascending from (7,500) to 15,000 feet *Juniperus pseudo-sabina*, which is taken by some as synonymous to *Juniperus Wallichiana*, reaches a little higher elevation up to 15,000 feet, being fairly common like that of *Juniperus recurva* between 9,000 and 13,000 feet.   Dallimore and Jackson consider *Juniperus recurva* " a native of the Eastern Himalaya occurring in " Sikkim and Bhootan, and *Juniperus pseudo-sabina* " a native of the Himalaya from the Indus to Bhootan ".   But records in the Calcutta Herbarium show that both the species are uniformly distributed forming evidently mixed association in the eastern and the western Himalayas, the latter only attaining a little higher elevation than the former. *Juniperus macropoda* is mainly an inhabitant of the Western Himalaya and abundant in Afghanistan and Baluchistan extending up to Persia.   Its predominant growth is observed between 8,000 and 12,000 feet, though some record it from an elevation of (5,000) feet.   It very rarely ascends, as Kurz' specimen shows, as high as 19,000 feet, in Werangpap Teedong Valley.   *Cephalotaxus Mannii* is confined only to the Khasia hills, Assam, between 3,000 and 6,500 feet, while

---

[1] The altitudes in brackets are taken from literature.

Cephalotaxas Griffithii predominates over the Mishmi hills, Upper Assam and passes into the Burmese hill ranges. This species occurs evidently in higher altitudes than *Cephalotaxas Mannii*, ascending from 3,000 feet to 8,000 feet; whereas *Cephalotaxas Mannii* extends from 3,000 to 6,500 feet only. Parkinson considers *C. Mannii*, a native of Burma. But there is no record to confirm this view. The distribution of *Taxus baccata* in the Himalayan ranges may be considered as the eastern spur of its great belt of distribution spreading over the British Isles, Europe, Northern Persia and Algeria, and finally entering the Himalayan ranges of Afghanistan, Bhootan, Assam and Upper Burma. In the Himalayas it extends uniformly both over the Western and the Eastern mountain ranges ascending from 5,000 to 12,000 ft., exhibiting its dominance in between 6,000 and 12,000 ft. Dallimore and Jackson mention Bhootan as its eastern limit of extension, but the sheets of this herbarium illustrate its access further into the Chin Hills and Ruby mines in Upper Burma, and perhaps still further, up to Chino-Tibetan borders. Of the two typical species of Indian *Podocarpus*, *Podocarpus latifolia* may be considered to be originally growing on the lower hill-forests of South India, Assam and Burma. This is the only Peninsular Indian Conifer growing in the Anamalais, Coimbatore. *Podocarpus nerifolia* has a wider range of distribution ascending up to 5,000 ft., growing abundantly in the lower hill-forests of the Central Himalaya, East Himalaya, East Bengal, (Chittagong Hill Forests), Assam, the Andaman Islands, South Burma and Malay Peninsula extending down to the Sunda Islands. Its record from China suggests its northern limit of extension, if the Chinese specimens prove definitely to be wild. Its extension so far north raises the problem how has it crossed the Burmese, Siamese and Chino-Tibetan borders, and whether our further knowledge of the flora of this important area might reveal its presence in the Eastern ranges of Tibetan Burmese and Siamese hills as well. *Podocarpus cupressina* appears to be a Malayan conifer which has gradually spread over the hill forests of Burma. *Pinus excelsa* is evidently a North-West Himalayan species ranging from an elevation of 5,000 to 12,000 ft. It is abundant between 6,000 and 12,000 ft., although, sometimes ascending up to 12,500 ft., where it often forms a constituent of the conifer forests. Its occurrence in the Central Himalaya, Sikkim and Chumbi Valley might be a later introduction. Dallimore and Jackson remark, however, that it extends " eastward to Nepal ". J. D. Hooker on the contrary is of opinion that it is " absent in Central and N.-W. Kumaon and in Sikkim ". I confirm Hooker's view from the data at my disposal. Hooker's suggestion of its doubtful occurrence in

Macedonia may be supported by the tendency of this species predominating in the North-Western ranges of the Himalayas, especially over the outer ranges extending to the Kafiristan in Afghanistan. Further, the existence of this species in Greece hints at its fairly long belt of distribution from Southern Europe through Persia to the Himalayas. But this fact can neither be definitely established from the data at present available, nor definite reason be adduced to the cause of this disconnected distribution in Macedonia and then in the N.-W. Himalaya, due again to our want of sufficient knowledge of this large tract of the country lying across the N.-Western Himalaya to the European border. *Pinus longifolia* is mainly confined to the lower-hill ranges, descending to the valleys of the western and the eastern Himalayas, where it extends up to Bhootan. It ascends up to 6,000 ft., but it is abundant in its wild state of growth in between 1,000 ft. and 3,000 ft. On the North-West it is said to form an extensive association between 1,500 and 6,500 ft., rarely reaching up to 7,500 ft., and above. This pine has adapted itself to grow in the plains and some of the magnificent trees are frequently found growing in the gardens of Northern India where climatic conditions are more favourable. But good specimens of fairly large size are observed to grow as low as nearly the sea-level. Thus in the Royal Botanic Garden, Calcutta, where this species had been first planted as early as 1794 or earlier, a fairly large number of trees of good size is still found growing there. (See plate II). *Pinus Khasya*—the well known Khasya pine—is exclusively a native of the Eastern spur of the Eastern Himalayan ranges spreading over the Khasya, the Jaintia and the Naga Hills and extending to the hills or Upper Burma and the Shan Hills, where it appears to gain predominance in growth. It ascends from an elevation of 2,000 to 6,500 ft., rarely 7,000 ft., and above. Its occurrence in the Philippines, if it is in its wild state there, suggests perhaps a touch of Malayan element in it. *Pinus Gerardiana* is chiefly a species of the North-western flank of the Himalayas extending from the Punjab Himalaya to Afghanistan and Baluchistan ranging from an altitude of (5,000) to 10,000 ft., sometimes ascending 11,000 ft., rarely 12,000 ft., and above. *Pinus Merkusii*, the Tenasserim pine, as its name suggests, is a Southern Burmese species spreading over low hills and extending to Cochin China, the Malay islands and the Philippines. It is said to be one of the most common conifers of Siam and occurs from an elevation of (500) to 2,000 ft., rarely 3,500 ft., and above. *Cedrus Libani* var. *Deodara*—(*Cedrus Deodara* of *Loudon*) is evidently a native of the North-West Himalayan ranges and occurs extensively, between elevations of (3,500) and 8,000 ft., sometimes

reaching 10,000 ft., rarely ascending 12,000 ft. It extends to Afghanistan as well. Brandis considers Kumaon and Nepal specimens of this Conifer cultivated. *Picea Morinda* (to this is included *Picea morindoides* of Rehder) spreads over the mountains of the Western and the Eastern Himalayas extending from near Afghanistan, Chitral, Hazara, Kumaon, Simla, Nepal to Sikkim. Its altitudinal range is from (6,000) to 12,000 ft., and rarely above. It is very likely that the species has penetrated into the hill ranges of Bhootan and beyond into the far eastern spurs of the Himalayan ranges. But this demands exploration in the hills of Bhootan and mountains of Northern Burma, although, Griffith's specimen of *Picea morindoides* from Bhootan (as noted by Dallimore and Jackson) substantiates uniform continuity of distribution of *Picea Morinda* from Chitral and beyond in the west, to Bhootan and beyond in the east. Its association with *Tsuga Brunoniana* also suggests continuity of distribution of one and the same species in the west and the east Himalayas. The occurrence of *Abies spinulosa*— (*Picea morindoides*) in Bhootan, as reported by Griffith, again confirms the suggestion of uniformity of distribution in the west and the east Himalayas. *Tsuga Brunoniana* forms an important constituent of the conifer forests extending over the mountains from Kumaon, Nepal to Sikkim, where it is said to be abundant in the inner ranges. It is common between elevations of 6,000 to 10,000 ft., below silver fir forests sometimes ascending slightly higher (10,500 ft.). *Abies Webbiana* is indigenous to both the western and the eastern Himalayan ranges extending from Afghanistan to Sikkim ascending from (7,000) to 12,000 ft., though sometimes rising up to 13,000–14,000 ft., in altitude. *Larix Griffithii* is confined to the Eastern Himalaya, growing profusely from Nepal to Bhootan between elevations of 8,000 and 12,000 ft., attaining in Sikkim and Tibet slightly higher altitude.

The occurrence of *Abies, Pinus excelsa, Juniperus recurva, Cupressus torulosa* in the North Burma Hills in recent years is evidently due to these species extending beyond their limit of Eastern border of distribution or might have been introduced later. The presence of *Tsuga yunnansis* in the Burmese borderlands, as recently discovered by Parkinson from Chimali pass Pahluka, 8,000 to 12,000 ft., is due to this Yunan and S. W. Szechuen species crossing the Chino-Tibetan frontiers. The presence of *Libocedrus Potaninii* in the forest across Burmese borderlands is due again to this S. W. Chinese species descending further southwards. Griffith's record of *Cupressus pendula* in Bhootan is evidently a cultivated form, as *C. pendula* is a synonym of the well known Chinese species *Thuja orientalis* var. *pendula*. As regards wild confers of Ceylon there is no definite records available to

prove which species of conifers are indigenous to the island. Although there are at present fifty species of conifers under cultivation, it is doubtful if there is one purely native of Ceylon. Out of a dozen of Marquand's record of conifers collected by Captain F. Kingdon Ward from the Eastern Himalaya and Tibet in 1924, only four species— *Pinus excelsa, Tsuga Brunoniana, Abies Webbiana* and *Larix Griffithii* are wild, the rest appear to be introduced in Tibet.

Ludwig Rudolph in his Atlas der Pflanzen Geographie über Alle theile der Erde Berlin (1864) shows the distribution of Tannen (Abies) in the N.-W. Himalaya and Fichten (Pines) in the Eastern Himalaya. In the world's distribution of Conifer forests—as delineated in the American Maps recently published—the Conifer formation appears to be somewhat isolated, except one more or less continuous belt running along the subarctic and temperate zones. The presence of Conifers in the Central Europe, Mediterranean regions and a few in the mountains of Northern Persia, then in the Himalayan ranges, China, Japan and Siam hills suggests a rather long somewhat uniform belt of Conifers in the subtropical zone, gaining predominance in the W. Himalaya. The distribution of Conifers in Asia confirms this view to a certain extent as the temperate Himalayan belt of Conifers has been considered as the fifth of the five lines of distribution of conifers suggested by Pilger. This line runs along the high temperate mountain regions of the Western and the Eastern Himalayas, Eastern Tibetan borders and the high mountain ranges of the Burmese Himalayas, Chino Tibetan frontiers, Yunan and Szechuan, finally passing unto the Chinese and Japanese Hill ranges. Further investigations in the unexplored regions may elucidate this hypothesis.

I offer my sincere thanks to the Forest Officers of the different provinces of the Indian Empire, particularly to Mr. R. N. Parker, Forest Botanist, Dehra Dun, and Mr. C. E. Parkinson, Forest Botanist, Rangoon, Burma, for their kind co-operation in the preparation of this paper.

List of Wild Conifers in the Indian Empire noting the localities of their distribution. The list is arranged according to Hooker's Flora of British India (Coniferae) Vol. V, pp. 643–653, 1890.

Order Coniferae

Genus No. 1. CUPRESSUS *Linn.* * *G. P. 6.*
Species No. 1. CUPRESSUS TORULOSA *Don.*

---

* G. P. refers to Bentham and Hooker's Genera Plantatum, Vol. III. pp. 420–442, 1883.

Area of general distribution :—

N. W. Frontier Province ; N. W. Himalya ; the Punjab ; Kashmir ; [1] Central Himalaya ; Assam (probably cultivated) ; Wallichian sheet [2] 6046.

Area of detailed distribution :—

Quetta Burial Ground at the foot of the Mountains, Julalabad, Afgan, Griffith ; near the Farra Rud, Afghanistan, T. Walter Irvine, 1905 ; District Hazara, Kagrin valley, 13,600 feet, Inayet, 1896 ; Afghanistan, Griffith, 1863–64 ; N.-W. Himalaya, District Chamba (Pangi), Sural Valley above Chabi Got, 12,500 feet, Harsukh, 1899 ; Pangi, Dr. Stoliczka ; Chamba State, J. H. Lace, 1898 ; District Janarsai, 9,000 feet, J. F. Duthie, 1894 ; Dewangari Hills Ter, 1855 ; Lokanda Marama Limestone, Dr. Brandis ; Mussourie, 6,500 feet, W. T. Saxton, 1915 ; Mussourie, George King, 1869 ; Falconer, 1865 ; Jemmai, J. S. Gamble, 8,000 feet, 1898 ; Elysium Hill, Simla, 6,500 feet J. S. Gamble, 1877 ; Kulu, 1882 ; Chenab Valley, Madagraon, 10,000 feet, 1881 ; Simla, 7,000 to 8,000 feet, Hooker f. and Thomson ; Boolyar 8,000 feet, T. W. Forster, 1894 ; Bhagirathi, banks of rivers 6,500 feet, Dr. Schlich, 1883 ; Central Himalaya, W. S. Webb ; Cuma above Naini Tal, 1866 ; Assam, Manipur, Political Agent, 1914 ; Manipur S. N. Bal (probably cultivated) ; Wallichian sheet 6046A, 6046B, Eastern Kumaon and Himalaya, W. S. Webb.

J. D. Hooker limits the distribution of this species up to Chamba from Nepal, but its occurrence in the outer Himalayan ranges in the west, as noted by Dallimore and Jackson, suggests its extension beyond Chamba, Hazara and Afghanistan and our sheets here confirm this view.

Species No. 2. CUPRESSUS SEMPERVIRENS *Linn.*

Area of general distribution :—

Afganistan, North-West India, Wallichian sheet 6046C, 6041D.

Area of detailed distribution :—

Afganistan, Griffith Herbarium, Lemann, 1852 ; Northern division Dr. Cleghorn, (Planted) ; Poona, 1890 (cultivated) ; N. Bengal, Purnea,

---

[1] The whole of the Himalayan ranges including the hills of the Northern Burma is divided into the eastern and the Western Himalayas. The Central Himalaya merges into the ranges of mountains where the Eastern and the Western spurs meet at the centre, in and about Western Nepal.

[2] Among the localities of the Willichian sheet numbers, only those which have special reference in the volume of Wallichian sheets have been noted in detail. The localities of the numbers which have general distribution in the Himalayas have not been repeated in the list of detailed distribution.

Cultivated, Kurz, 1868 ; old Agartolah, Tippera Hills, 5,000 to 8,000 feet, P. M. Debbarman, 1914 (planted) ; Burma, Maymyo, plateau, 3,500 feet, C. G. B. Dawkins ; Sadon, 4,500 feet. Lady Cuffe 1915, (Hooker mentions this as cultivated). [This is an introduced species which has acclimatized itself in the North-West India earlier than 1852.]

Species No. 3. CUPRESSUS FUNEBRIS *Endl.*

Area of general distribution :—

Nepal ; Sikkim ; Bhootan.

Area of detailed distribution :—

Dehra Dun, 2,200 feet, J. S. Gamble, 1894 (cultivated) ; Sikkim, Yaksum, 4,500 feet, T. Anderson, 1862 ; Tista River, between Darjeeling sub-division and border point of Sikkim, Dr. Schlich, 1873 ; Sikkim, 5,000 feet, Hooker f., and Thomson, Sikkim Himalaya, Dobdie Monas-tery, Toukson, G. Watt, 1887 (cultivated) ; East Himalaya-Griffith, 1861–1862. [This species is also, according to Hooker, grown near Buddhist temples in Nepal, Sikkim and Bhootan. This species, a native of Central China, was introduced, and established itself in this country before 1860.]

Species No. 4. CUPRESSUS LUSITANICA, *Mill.* var. *Benthamii.*

Area of general distribution :—

N.-W. Himalaya.

Area of detailed distribution :—

N.-W. Himalaya, Kaulagarh Tea Estate, Dehra Dun, R. N. Parker, 1922, (probably cultivated). This is a native of Mexico, introduced in this country in early days.

Species No. 5. CUPRESSUS CASHMIRIANA *Royle.*

Area of general distribution :—

East Himalaya.

Area of detailed distribution :—

East Himalaya, W. E. Smith, cultivated in N. Italy and supposed to be identical with the Bhootan specimens collected by Griffith at Dewangiri.

Species No. 6. CUPRESSUS SP. (unidentified).

Area of general distribution :—

N.-W. Himalaya.

Area of detailed distribution :—

Dist. Chamba, Pangi Sahaul Road, near Salgraon, 9,000 ft. ; Harsukh—1899, (may or may not be cultivated).

Genus No. 2. JUNIPERUS LINN. *G. P. 7.*

Species No. 1. JUNIPERUS COMMUNIS STATE *Linn.*

Area of general distribution :—

Afghanistan N.-W. Himalaya ; Wallichian sheet 6044.

Area of detailed distribution :—

Afghanistan, Ballad; Gilgit expedition, Hindukush, Dr. Giles, received through Mr. Duthie, 1887; Kashmir, Suknullah, Dua's valley, 11,000 ft. to 12,000 ft.; Kashmir, above Doyen, Dist. Astor and Karachu valley (1891), 12,000 ft. to 13,000 ft., J. F. Duthie, 1892; N.-W. Himalaya, Chamba (Pangi), Ajog forest, 8,500 ft., Harsukh, 1889; Tola Kumaon-Himalaya, 11,500 ft., R. Strachey and J. E. Winterbottom; Kuti, Kumaon, 11,000 ft., S. R. Kashyap, 1926, Var. *Alpina*, Millum, Kumaon, R. Strachey and J. E. Winterbottom, 1848; Chamba State, N. W. Himalaya above Kilar, 10,000 to 12,000 ft., J. H. Lace, 1896; N.-W. Himalaya, Tehri Garhwal, Chensil range, 11,000 ft., J. F. Duthie, 1894; Jambatai, Chitral, 10,660 ft., Surg. Lt. Harriss, 1895; N.-W. Himalaya, Hazara, Kagun valley, 13,600 ft., Inayet, 1896; Pangi (Lahoul), Chamba State, 10,000 ft., J. H. Lace, 1897; Exalpibus Himalayana, Dr. W. S. Webb; Himalayas, George King (cultivated), 1869; N.-W. Himalaya, Kibar dogri, 9,000 ft., J. H. Lace, 1890; W. Himalaya, 5,000 to 11,000 ft., Hook. f. and Thomson; Parbanee 9,000 to 9,500 ft., Dr. Brandis, 1864; N.-W. Himalaya, Roghie, Dr. Brandis; Lahoul, 10,000 ft. to 12,000 ft., R. W. Heyde, 1877; Kashmir, Kangan, Sind valley, 5,500 ft., G. A. Gammie, 1891. Wallichian sheet 6044–1824, Neetee, W. S. Webb, East Srinagar up to Kumaon India orientalis, Dr. Wallich. 1869.

Species No. 2. JUNIPERUS PSEUDO-SABINA *Fisch. and Mey.*

Area of general distribution :—

N.-W. Himalaya; Tibet; Nepal; Sikkim; Chumbi; Bhootan; Wallichian sheet 6041A, B. C.

Area of detailed distribution :—

Chamba, Pangi, Near Triloknath, 8,000 to 12,000 ft., J. H. Lace, 1897; Kashmir Himalaya, Kilane, Kumaon, 11,500 ft., R. Strachey and J. W. Winterbottom, 1848; Kuti, Kumaon, 13,000 ft., S. R. Kashyap, 1926; Srinagar, Robert Brown; Tehri Garhwal, Gangotri, 12,000 ft. to 13,000 ft., J. F. Duthie, 1881; Chamba-Comm. Robert Ellis, 1880; W. Tibet, Gode in Hasara (Belli or little Tibet Chumbi, Neepan and Kashmir, J. E. Winterbottom, 1847; Nepal, Dr. J. Scully; Sikkim—Himalaya, King's collector, 1888; Sikkim, 10,000 to 15,000 ft., Teumthang, G. King, 1885; Toumrachen Chu, 12,500 ft., Smith and Cave 1909; Sikkim, 12,000 ft., W. Wallich, 1870; Ratong, Sikkim, 1857; Tongri, Sikkim Tougpong 13,000 ft., G. Watt. 1881; Tongri, T. Anderson, 1862; Phalloot, Sikkim, 13,000 ft., S. Kurz; Tey lep Pau 13,000 ft., J. S. Gamble, 1880; Sikkim subalpine, 10,000 to 15,000 ft., Hook. f. and Thomson; West of Jongri, 12,500 ft., G. A. Gammie; Chumbi-Kimpau, 1877; Chumbi State, Kukti pass, J. H. Lace, 1897; Rashorg

valley, 10,000 ft., Tolling Hills, Dr. Brandis; Bhootan, Dr. King's collector, 1888; Bhootan hills, H. Hamilton; Kishtwar—Subalpine, Hook. f. and Thomson; Himalaya Tarlaria confines W. S. Webb, Srinagar, Gosiathan, Wallichian sheet 6041.

J. D. Hooker reduces *J. Wallichina* to *J. pseudo-sabina*, but Dallimore and Jackson, who consider Hooker as the author of *J. pseudo-sabina* and not Fischer and Meyer, have reduced *J. pseudo-sabina* to *J. Wallichiana* Hooker f.

Species No. 3. JUNIPERUS RECURVA *Ham.*

Area of general distribution :—

N.-W. Himalaya; Chitral; Tibet; Nepal; Sikkim: Chumbi; Bhootan; Wallichian sheet 6042 A.B.

Area of detailed distribution :—

N.-W. Himalaya, Chitral Relief Expedition, Lawari Pass, 10,500 ft., Brig. Genl. Gatacre D. S. O., 1895; Hazara, Siran Valley, Inayet, 1896; Kashmir, J. F. Duthie, 1893, var. *Squamata*, Parlatore, Wallichian sheet 6043 C., Alpine Himalaya; Srinagar, R. Brown; N.-W. Himalaya, Dr. Stoliczka; Horang, Dr. Brandis, 10,000 ft., J. F. Duthie; N.-W. India, H. B. Royle; Tehri Garhwal, Kidarkanta, 10,000 ft., to 11,000 ft. J. T. D. 1879; Intul 12,000 ft., Dr. Schlich, 1883; [1] N.-W. Himalaya, Lahaul, 10,000 to 12,000 ft., Heyde; Chitral Expedition, Bundai, 9,600 ft., Surg. Lt. Harriss, 1895; Lawripass, Chitral Expedition, 10,500 ft., Brig. Genl. Gatacre, 1895; Sutlej valley, S. R. Kashyap, 1923; Samada, Central Tibet, on the road to Gyantse, 14,100 ft. Way to Kupup 13,000 ft., S. R. Kashyap, 1929; Pindaree glacier, Kumaon, 12,500 ft., Sunder dunga 12,000 ft., Kumaon, Niti, 11,500 ft. Garhwal, R. Strachey and J. E. Winterbottom, 12,000 ft., 1848; Tolu Kumaon—11,500 ft., R. Strachey and J. E. Winterbottom, 12,000 ft., 1848; Margraon, J. H. Lace, 1897; Yatung, S. R. Kashyap, 1930; Khambajong Tibet Frontier Commission, Kajor. F. E. Younghusband, 1903; Nepal, Dr. J. Scully; East Himalaya and Gasaithan, Wallich, 1824; Sikkim Himalaya Yeumtong (Lachung valley), 13,000 ft., Sibu valley, 12,000 ft., G. A. Gammie, 1892; Guatong, 11,000 ft., W. Wallich, 1874; Sikkim, 10,000 to 12,000 ft., Hook. f. and Thomson; Dr. King's collector, 1887; Singalelah, Darjeeling, 10,000 ft., C. B. Clarke, 1870; East Himalaya, Griffith, 1861–62; Bhootan, Sergea mountain, summit of ledge towards Rydams, 10,000 ft., ascent of hill to Rydam 9,500 ft. to 10,000 ft. Wallichian sheet 6042 A.B. 6043 ex Himalaya, 1824.

J. squamata, Ham. is taken as an independent species by Dallimore and Jackson. According to Clinton and Baker this species

---

[1] Dr. Schlich's collection is supposed to be a variety, var. *squamata* Parlat.

differs from *J. recurva* in having stouter and broader leaves and smaller and slightly different kinds of fruits. Its occurrence, as two separate species—*J. recurva* and *J. squamata* in Nepal has been recorded by D. Don as well.

Species No. 4. JUNIPERUS MACROPODA *Boiss.*

Area of general distribution :—

Afghanistan ; Baluchistan ; N.-W. Himalaya ; Chitral ; Trans-Indus Himalaya ; West Tibet ; Assam ; Wallichian sheet 604/A.

Area of detailed distribution :—

Rawai and Hazara, Afghan ; Quetta, 1909 ; Ghushki, 8,000 ft., J. H. Lace, 1886 ; Rarang, 10,000 ft., J. H. Lace, 1890 ; Milum, Kumaon, 11,500 ft., R. Strachey and J. E. Winterbottom, 1848 ; Kashmir, 6,700 ft., T. Thomson ; Chamba, Punjab, 10,000 ft., A. Pengelly 1887 ; Simla, Nilany, 10,000 ft. to 11,000 ft. up to 12,000 ft.; Dr. Schlich, 1883 ; N. W. Himalaya ;Talling Hills and Chargo, Dr. Brandis, Drankar, 12,000 to 14,000 ft., Dr. Stoliczka ; Werangpap Teedong valley ; 19,000 ft., S. Kurz ; Kashmir, 11,000 to 12,000 ft., Astor, 8,000 ft., 1892 ; J. F. Duthie, 1892 ; Jeolikota, Kumaon, N. Gill, 1913 ; Lahul, Jispa, S. R. Kashyap, 1919 ; Chitral Relief Expedition 11,000 ft., Surg., Lt. Harriss, 1895 ; Ba—N. W. Tibet, 8,000 ft., T. Thomson ; Tibet, 5,000 to 15,000 ft., T. Thomson ; Manipur, Assam, Political Agent, 1914 ; (probably introduced in Assam, as *Pinus Khasya* is evidently the only conifer found growing wild in the Naga Hills and Manipur area).

Genus No. 3. CEPALOTAXUS, *Sieb. & Zucc. G. P. 12.*

Species No. 1. CEPHALOTAXUS MANNII *Hook. f.*

Area of general distribution :—

Khasia Hills ; Assam.

Area of detailed distribution :—

Khasia Hills, 5,000 to 6,000 ft., Muplong, and Lankhla woods, 3,000 ft., G. Mann, 1885 ; Shillong, Jowai Road, Dr. Prain, 1892 ; Bernardungo, 6,500 ft., J. W. Oliver, 1892 ; Themokidima Forest, Assam, 5,000 ft., G. Watt, 1895.

Species No. 2. CEPHALOTAXUS GRIFFITHII *Hook. f.*

Area of general distribution :—

Assam ; Burma.

Area of detailed distribution :—

East Bengal, 5,000 ft., Griffith ; Duphla hills, J. L. Lister, 1874 ; Above Konoma, South East of outpost about 8,000 ft., Dr. Prain, 1886 ; Naga Hills, High range of hills, near the sources of Kapila river, North Cachar, Capt. Bewar, 1857 ; Manipur, G. Watt, 1881–82 ; Saunta-

thonlon, Burma, 3,000 to 4,000 ft., A. Rodger, 1916; Upper Burma, J. T. W. Leslie, 6,000 ft., 1890; Burma, Ruby mines, J. W. Oliver, 6,500 ft.

Dallimore and Jackson, however, mention that this species occurs in Mishmi Hills, Upper Assam, at an elevation of 6,000 ft. Griffith's "East Bengal" evidently refers to Eastern Himalaya including Assam. *Species* No. 3. CEPHALOTAXUS BACCATA, Linn, as noted on one of the sheets of the Genus *Cephalotaxus* has been collected from Mongnai, Southern Shan States, by W. H. Craddock in 1900 and mentioned on the sheet "found in wild state" appears to be a form of *C. Griffithii*. I have not been able to trace this specific name in the literature available in Calcutta.

Genus No. 4. TAXUS *Tournef.* G. P. 13.

Species No. 1. TAXUS BACCATA, *Linn.*

Area of general distribution :—

Afghanistan; N.-W. Himalaya; Nepal; Sikkim; East Himalaya and Assam: Upper Burma; Wallichian sheet 6054 A, B, C, D, E. 484, 6,055.

Area of detailed distribution :—

Afghan., 1857; W. Himalaya-Hazara dist., 5,000 ft., and above, Inayet, 1899; North-West Frontier Province Janusar, Deoban 9,000 ft., F. W. Forster, 1894; Temperate west Himalaya, Hook. f. and Thomson, 1877; Jawnsar divs., Mundali dist., Simla, Hirasingh, N.-W. Himalaya, J. K. Knowles, 1920; Kumaon, 8,500 ft., Jegeswar, 1848; below Baling, Kumaon, 10,000 ft., S. R. Kashyap, 1926; Simla, 9,000 ft., Gamble; N.-W. Himalaya, Tehri Forest above Deota, 8,900 ft., J. F. Duthie, 1895; Chamba State, Kalai Forest, J. H. Lace, 8,000 ft., 1899; Urni Forest, 9,500 ft., J. H. Lace, 1890; Chamba Robert Ellis, 1880; Mulluk and S. of Bhabel, 9,000 to 12,000 ft., Dr. Stoliczka; Nachar Forest, Bursahir, Dr. Brandis; 8,000 ft., Nepal, Wallich, 1821; Webb, R. B. and others, 1822, 1824; Sikkim, Kurz, Hook. f. and Thomson, 7,000 to 10,000 ft., H. D. Hooker; Sangloo, Sikkim, 8,000 to 10,000 ft., T. Anderson, 1862; Tangloo, C. G. Roger, 1899; East Himalaya, Griffith, (Griffith notes *Taxus sp.*, from Bhootan collection as well) 1861; Khasia Hills, Assam, in the *Quercus Rhododendron* wood, Muplang, H. G. Carter, 1920; Temperate region, 5,000 to 6,000 ft., Hook. f. and Thomson; Khasi hills and Brahmaputra plains, Kurz; Wallichian sheet, Khasia Hills, 1850; Khasia and Jaintia Hills, 5,000 ft., Nungbiai, G. Mann, 1855; Japoo, Manipur, 1882; Japoo, Manipur, 8,000 ft., G. Watt, 1881–82; 7,000 ft., Manipur, G. Watt, 1882; Chin Hills, Upper Burma, C. R. Dun, 1895; Burma, Ruby mines, 6,500 ft., T. W. Oliver.

Genus No. 5. PODOCARPUS *L. Herit. G. P. 21.*

Species No. 1. PODOCARPUS LATIFOLIA *Wall.*

Area of general distribution :—

Assam ; Burma ; S. India ; Wallichian sheet 6050.

Area of detailed distribution :—

Assam, G. Mann, 1893 ; East Bengal, Griffith, 1863–64 ; Mt. Sillet, De Silva ; Barakres 2,500 ft., Kanjilal, 1914 ; King's collector 1893 ; Shillong, G. Mann, 1887 ; Tavoy, Burma, 1925 ; Pegu-Burma, Kurz ; Moulmain, Falconer, 1849; South India, C. A. Barbar, 1908 ; Anamalais, S. Coimbatore 4,000 ft., C. C. Wilson, C. E. C. Fischer ; Wallichian sheet 6050.

Dallimore and Jackson consider *P. latifolia Wallich* as a synonym of *P. Wallichianus C. Presl.*

Species No. 2. PODOCARPUS NERIIFOLIA, *Don.*

Area of general distribution :—

Central Himalaya ; East Himalaya ; East Bengal ; Assam ; Andamans ; Burma ; Malaya Peninsula ; Wallichian sheet 6052 A, B & C.

Area of detailed distribution :—

Nepal, Dr. Wallich, 1889 ; Bhootan, Debrapur, 1864 ; East Himalaya, Sikkim, 3,000 ft., T. Thomson, S. Kurz ; Gangtak, Riboo and Rhompo ; East Bengal, Griffith, 1863–64 ; Assam, Abor Expedition, above Balek, 2,300 ft., I. H. Burkill, 1911–12 ; Khasia Mt., Oldham ; Khasia, 2,000 to 3,000 ft., Hook. f. and Thomson ; Khasia Hills, Sylhet, Jaintia Hills, 5,000 ft., G. Mann, 1886 ; Assam, Nambur Forest, G. Mann, 1891 ; Chittagong Hill Tracts, J. S. Gamble, 1880 ; J. S. Lister, 1876 ; Andamans, King's Collector, 1884 and 1890 ; South Andaman, S. Kurz ; Burma, Maymyo, Maung Kan, 1924 ; Tenasserim, G. Gallatly, 1877 ; Wallichian sheet 6052 A, B, C, Nepal, Singapore, Calcutta Botanic Garden, 1822.

Species No. 3. * PODOCARPUS CUPRESSINA *Br.*

Area of general distribution :—

Burma ; Malay Peninsula.

Area of detailed distribution :—

Hupung Valley, Burma, J. Wallace, 1856.

Species No. 4. PODOCARPUS WALLICHIANUS *C. Presl.*

Area of general distribution :—

Malay Peninsula ; Wallichian sheet 6057.

Area of detailed distribution :—

Penang, Wallich, 1822 ; Perak, Scortechini ; Goping Kinta, L. Wray, 1883 ; Bakit saga, State of Johor, 1890.

This species is nothing but different forms of *P. latifolia* Wallich and I agree with Dallimore and Jackson in reducing *P. latifolia* of Wallich to *P. Wallichianus.*

Genus No. 6. PINUS *Linn. G. P. 26.*

Species No. 1. PINUS EXCELSA *Wall.*
Area of general distribution :—

Afghanistan ; N.-W. Himalaya ; Chitral ; Nepal ; Chumbi ; Wallichian sheet 6059 A, B, C, 2670, 307, 1821–1824.
Area of detailed distribution :—

Afghan ; N.-W. Province, Boolyar, T. W. Forster, 1895 ; Jehru, N.-W. Province, 8,000 ft., J. S. Gamble, 1891, Junswar, 7,500 ft., J. S. Gamble, 1894 ; Junswar, 8,000 ft., J. F. Duthie, 1898 ; N.-W. Himalaya, Dr. Stolickza ; Dr. Brandis ; Bashahr, N.-W. Himalaya, Bahli, 7,500 ft., J. H. Lace, 1890 ; Naldehra, N.-W. Himalaya, J. S. Gamble, 1878 ; Simla, J. S. Gamble, 1877 ; Kashmir, 5,000 to 11,500 ft., Hook. f. and Thomson ; Chamba, N.-W. India, Robert Ellis, 1880 ; Mussourie, G. King, 1869 ; Tehri Garhwal, Lambatach, 7,000 ft., J. F. Duthie, 1897; Chitral Expedition, Surg. Lt. Harriss, 1895 ; Murree Hills, Upper Topa, 6,800 ft., T. A. Sprague, 1910 ; Sikkim, cultivated, 6,000 to 10,000 ft. J. D. Hooker ; Chumbi, J. W. Edgar and Dingboo, 1877 ; East Himalaya, Griffith ; Assam, Col. Jenkins (perhaps introduced). [There is no Herbarium sheets from Nepal available here, but David Don reports its occurrence from Nepal basing evidently on Hamilton and Webb's collection of this species. Marquand records this species from Kingdon Ward's collection of E. Himalaya and Tibet, from Tsang-Po Gorge 2,100 to 2,400 m.]

Species No. 2. PINUS LONGIFOLIA *Roxb.*
Area of general distribution :—

N.-W. Himalaya ; Wallichian sheet 6065 A and B, 1861.
Area of detailed distribution :—

N.-W. Himalaya, G. King and Brandis ; Murree Hills, Lower Topa, T. A. Sprague, 1918 ; Nurpur-Kangra Dist., I. H. Burkill, 1902 ; Jaru, Simla, 6,000 ft., J. S. Gamble, 1877 ; Sikkim, J. D. Hooker ; Bhootan, W. sheet Nepal, Horto. Botanico, Calcuttensis (evidently cultivated) 1861.

Species No. 3. PINUS KHASYA *Royle.*
Area of general distribution :—

Khasia Hills ; Assam ; Burma ; Wallichian sheet 37499 A. 6064 A.
Area of detailed distribution :—

Khasia and Jaintia Hills, Mann ; East Himalaya, Rungeet, 2,000 ft., G. H. Cave ; Assam, Jenkins ; Shillong, 5,000 ft. C. B. Clarke, 1885 ;

Kohima, Naga Hills, Dr. D. Prain, 1886; East Bengal, George Watt 1881–82; Inle Lake, Southern Shan States, Burma, N. Annandale, 1917; Upper Burma, C. R. Dun, 1895: Nut-toung Mts., Burma, Cross, 1861; Burma, Dr. Brandis; Upper Burma, Ruby Mines, Abdul Huk, 1891; Burma, 4,500 ft., J. M. D. Mackenzie, 1915; Koni, Upper Burma, J. C. Prazer; Burma, Pegu, Broke Ridge, S. Kurz.

Species No. 4. PINUS GERARDIANA *Wall.*

Area of general distribution :—

Baluchistan; N.-W. Himalaya; Chitral; Wallichian Sheet 6064.

Area of detailed distribution :—

British Baluchistan, Barsukh (Afghan) 1897; Bashahr, N.-W. Himalaya, Kilbato Sholta, 6,000 ft., J. H. Lace, 1891; Gilgit, Dr. G. M. Giles, 1885; Astor valley, 8,000 ft., J. F. Duthie, 1892; Punjab, Himalaya, Pangee, Dr. D. D. Cunningham, 1884; Kunawar, 6,000 to 10,000 ft., Hook. f. and T. Thomson; Dr. Stoliczka; Chitral Expedition, 10,000 ft., Surg. Lt. Harriss, 1895.

Boissier records in his Flora Orientalis Aitchinson's collection of this species from "Hariab et Kost Affgheniae orientalis 7,000 to 11,000 ft."

Species No. 5. PINUS MERKUSII *Jungh. and De Vriese.*

Area of general distribution :—

Burma (Upper Tenasserim) ; Malaya ; Siam.

Area of detailed distribution :—

Burma, Morgui Dist. Maungook peak, 3,500 ft., Gilbert Rogers, 1910 Amherst Dist., Thaungyin valley, J. H. Lace, 1909; Shan States, 1,700 to 5,000 ft., J. H. Apbin, 1887; Martaban, Thoungyen, Dr. Brandis.

Pinus montana Dursi, cultivated in Nepal; *P. Laricio Poir.*, introduced in Simla and N.-W. Himalaya and cultivated there, collected in 1919; *P. Pinaster*, Soland, grown in the Government orchards, Ranikhet, U. P. Wallichian sheet—7278, Nepal; *P. sylvestris* introduced and cultivated in Ranikhet—U. P. 1920; in South India and Ootacamund in 1857 and 1858. The above species of *Pinus* have more or less adapted themselves to the climatic and edaphic conditions of this country.

Genus No. 7. CEDRUS *Laudon. G. P. 27.*

Species No. 1. CEDRUS LIBANI *Barrel., var.* DEODARA *Hook. f.*

Area of general distribution :—

N.-W. Himalaya; Gilgit; Kumaon; Wallichian sheet 23286 D, 6060 A, B, 1821.

Area of detailed distribution :—

Afghan, Strachey, 1857; N.-W. Himalaya, Hazara, 4,500 to 9,000 ft., Stewart; Junswar Division, 8,000 ft. J. F. Duthie, 1894,

J. W. Forster, Dr. Brandis; Pangee, Dr. Stoliczka; Chitral Relief Expedition, 7,000 ft., Surg. Lt. Harriss, 1895; Dalhousie, 7,000 ft., C. B. Clarke; Simla, T. Thomson, 7,000 to 8,000 ft., Dr. Schlich; Dehra Dun, U. P., Mundali, 8,000 ft., B. R. Bade, 1909; Deoban, 8,500 ft., A. V. Kesaviengar, 1908; Kumaon, Garhwal, 1857; (Tehri Garhwal) 10,000 ft., Duthie; Nepal, cultivated, 1884; cultivated in Khasia hills, 4,000 to 6,000 ft., Aitchinson's collection of this species from the mountains of Dist. Kuram Affghaniæ orientalis 7,500 to 10,000 ft., as noted by Bossier may also be mentioned here. Wallichian sheet Nos. 6060 A, Kumaon, R. B., Nepal 1821.

This variety of Barrelier's C. Libani has been raised by Loudon to the rank of the species *Cedrus Deodara* Loudon — as held by Dallimore and Jackson; and they have sufficient justification in considering C. Libani, Barrelier; (Cedar of Lebanon) as a separate species.

Genus No. 8. PICEA, *Link. G. P. 28.*

Species No. 1. PICEA MORINDA *Link.*

Area of general distribution :—

N.-W. Himalaya; Sikkim; Wallichian sheet 6063.

Area of detailed distribution :—

Baklidhar, J. H. Lace, 1891; N.-W. Himalaya, Dr. Brandis; Nirrkhamla, Dr. Schlich, 1883; W. Himalaya, 7,000 to 9,000 ft., T. Thomson; Hazara, Kagaon valley, 9,000 to 5,000 ft., Inayet 1896; Chamba State, Gothan ridge, 8,000 ft., J. H. Lace, 1898; Jaunsar, 8,000 ft., J. F. Duthie 1893; Chitral Expedition, Guger, 10,000 ft.; Surg. Lt. Harriss, 1895; Simla, J. S. Gamble, 1877; Kumaon, N. Gill, 1913; Mussourie, G. King; Imperial Forest College, Dehra Dun—U. P., Mundali, 8,000 ft., B. R. Bade, 1909; N.-W. Himalaya, Droban, Dr. Brandis; Sikkim, Lachung 9,000 ft., G. A. Gammie, 1892; Lachen, 10,000 ft., King's collector, 1885; Yeumthong, 12,000 ft., G. H. Cave, 1915; King 1875–76; (*Picea morindoides* Rehder), Zemu valley, 8,500 ft., Smith and Cave, 1909; Sikkim, 8,000 to 10,000 ft., J. D. Hooker, 1885; Chumbi, 12,000 ft., J. S. Gamble, 1880; Ha-ulong-pg-ong, King's collector, 1884; Chumbi Phari, Dungboo, 1879.

Griffith reports the occurrence of *Abies spinulosa*, which is synonymous to *Picea Morinda* Rehder, in Bhootan. Rehder's *Picea morindoides* is evidently, as Dallimore and Jackson remark, perhaps with reference to Troup and other authorities, a "more tender form than *P. Morinda*". This may be due to edaphic and climatic variations as considerable variations are noticed among the Herbarium specimens. Hooker's *Picea Morinda* must have included these tender forms as well, and I doubt how far the separation of these tender forms of *P. Morinda* raising them to the rank of a species is justified. I have, therefore, considered (after Hooker) *P. Morinda* as the only species

including this tender form (*Picea morindoides*), which is predominant in the Sikkim Himalayas. Evidently on this ground the sheet identified as *P. morindoides* collected by Smith and Cave from Zemu valley has been kept in the bundle of *P. Morinda*. There is, however, following remarks on one of the sheets marked *Picea Edgeri* "*Picea morindoides*—flat-leaved spruce—Chumbi valley. Griffith—Bhootan, Hooker—Lachen, Hooker—Yetung. Question is, does *Picea Morinda* occur at all in Sikkim or E. Himalayas. We have no specimen of Kew of it from them."

Genus No. 9. Tsuga *Carriere. G. P. 29.*

Species No. 1. Tsuga Brunoniana *Carr.*

Area of general distribution :—

N.-W. Himalaya ; Nepal ; Sikkim ; Chumbi Bhootan ; Wallichian sheet 6055, 6061, 1824.

Area of detailed distribution :—

N.-W. Himalaya ; Kumaon, Shosa Kali valley, Inayet, 1900 ; Kumaon, 10,000 ft., J. F. Duthie, 1884 ; Nepal. Dr. J. S. Scully ; Wallichian sheet 6061, Nepal 1824, Sebu valley—Sikkim, 10,000 ft., G. A. Gammie, 1892 ; Jemu valley, 9,000 ft., Smith and Cave, 1909 ; Zeumthong, 11,000 ft., G. H. Cave, 1915 ; Sikkim 8,000 to 10,000 ft., J. D. Hooker ; Phalloot descent, 11,000 ft., S. Kurz ; Lachung, 1883 ; Chumbi and Phari, Rinchingong, Dunboo, 1878 ; Chumbi, Neempen ; Ta-ssie-chen-loom, Chumbi, King's collector, 1884 ; East Himalaya, Griffith, 1861–62.

Marquand records this species from Kingdon Ward's collection of E. Himalaya and Tibet in Tsangpo Gorge forest above Gompo Ne.

Engler in his 2nd Edition of Naturlichen, Pflanzen Familien, 2 Auflage, 13 band, P. 186, 1926, remarks that *Tsuga Brunoniana* occurs in the inner ranges of the mountains of the Eastern Himalaya from Nepal to Bhootan. Hooker, however, mentions that it is wild in the Temperate Himalaya from Kumaon to Bhootan, between 8,000 to 10,500 ft. The localities noted on the sheets of the Herbarium specimens available in the Calcutta Herbarium confirm Hooker's statement. Don's *Pinus dumosa* occurring in Nepal is a synonym of *Tsuga Brunoniana*, so also *Pinus Brunoniana* of Wallich.

Genus No. 10. Abies *Juss. G. P. 31.*

Species No. 1. Abies Webbiana *Lindley.*

Area of general distribution :—

Afghanistan, Trans-Indus-Himalaya ; N.-W. Himalaya ; Nepal ; Sikkim ; Assam ; Wallichian sheet 6063, 6056, 1824, 6058A, 6060 A. H.

Area of detailed distribution :—

Afghanistan, Griffith, 1852 ; Bashahr, Baklidhar, 9,000 ft., J. H. Lace, 1891 ; N.-W. Himalaya, Mussourie, G. King, 1869 ; N.-W

Himalaya and Kumaon, Hazara, Dr. Brandis; Chamba, N.-W. India C. R. Ellis, 1880; (var. *Pindrow*) below Bahing Kumaon, 10,000 ft., S. R. Kashyap, 1926; N.-W. Himalaya, S. Kurz; Royle, Deoban, N.-W. Himalaya, Dr. Brandis; 9,000 to 12,000 ft., T. Thomson; Dehra Dun, U. P. Deoban, 8,500 ft., A. V. Kesaviengar, 1908; Above Dwali, 9,500 ft., Kumaon, R. Strachey and J. E. Winterbottom; Kathi Pap, 9,000 ft., R. Strachey and J. E. Winterbottom; Junsai 8,000 ft., J. S. Gamble, 1895; Tehri Garhwal, 13,000 ft., J. S. Gamble, 1893; N.-E. Himalaya, Kumaon, T. Anderson, 9,000 ft., 1857; Nepal, Dr. J. Scully; Wallichian sheet, 6058 A, Gosiathan, 1821; Sikkim, G. A. Gammie, 1892; Sandakphu, G. A. Gammie; Sikkim, 10,000 to 12,000 ft., J. D. Hooker; Phalloot, 10,000 ft., 1887; Sikkim, S. Kurz, 1868; Dungboo, 1878; Watt, 1881; King's collector 1882 and 1884; Thumku, 12,000 ft., King's collector, 1885 Assam, Jenkins.

Marquand records this species from Kingdon Ward's collection of E. Himalaya and Tibet. Tsang-Po Gorge, 3,000 to 3,400 ft., 1928.

Genus No. 11. LARIX *Miller, G. P. 32.*

Species No. 1. LARIX GRIFFITHII *Hk. f. & Th.*

Area of general distribution :—

Nepal; Sikkim and Bhootan.

Area of detailed distribution :—

Sikkim, Jemu valley, 9,000 ft., Smith and Cave 1909; Sikkim, Lachung, 10,000 ft., Smith and Cave 1909; 9,000 to 12,000 ft. G. A. Gammie, 1891; Dr. Cunningham, 1889; Lachen, 10,000 ft., King's collector, 1885; Yeumthang, 11,000 ft., G. H. Cave, 1915; 11,000 ft., J. D. Hooker; Chumbi, Dungboo 1877, 1878, 1879; East Himalaya, Griffith; Jutse to Phari, S. R. Kashyap, 1929; Below Chumpithang and Yatung S. R. Kashyap, 1930.

Marquand records this species from Kingdon Ward's collection of E. Himalaya and Tibet, Tsang-Po valley, Tibet, 3,400 to 4,000 m. 1928.

Hooker notes that this species is confined to Eastern Nepal, Sikkim and Bhootan, altitude 8,000 to 12,000 ft. Pilger in the new edition of Pflanzen Familien reports its presence at as low as 2,700 m. It appears from the sheets available in this herbarium that there is no record from Burma. Mr. C. E. Parkinson considers this too as a native of Burma. I have not yet received any Burmese specimen to confirm Parkinson's statement. Its occurrence might be due to its extension in the Burmese hill ranges in later years.

Genus No. 12. *DACRYDIUM *Soland. G. P. 17.*

Species No. 1 *DACRYDIUM ELATUM *Wall.*

Area of general distribution :—

Malay Peninsula; Sumatra; Java; Fiji; Singapore; Wallichian sheet 6045.

Area of detailed distribution :—

On Mount Ophir, Malacca, A. C. Maingay, 1867; Penang, Dr. Stoliczka planted in the garden on top of Penang Hill, G. King, 1879; Fiji island, Dr. Suman 1860; Gunong Tahan, Pahang, L. Wray and H. C. Robinson, 1905; Gunong, Bubu Larut, 4,500 ft., L. Wray 1890; Penang, Dr. Wallich; Singapore, Sir R. Schomburgh 1859; *D. falciforme* Pilg., Malay Peninsula. Kurz records its doubtful occurrence in Tennaserim.   D. Beccari Paul, Malay Peninsula; Wallichian sheet 6045, Penang Jack and Wallich 1819, 1822, Is. Phillippines, Wallich 1824.

Genus No. 13. *AGATHIS *Salisb. G. P. 23.*
Species No. 1. *AGATHIS LORANTHIFOLIA *Salisb.*
Area of general distribution :—

Malay Peninsula; Wallichian sheet 6037 A.
Area of detailed distribution :—

Waterfall Hill Larut Perak, 2,500 ft., G. Wray; Government Hill, Penang, A. C. Maingay, 1867; Royal Botanic Garden, (cultivated) 1915; Botanic Garden, Calcutta, 1834; Collected from Hort. Bot. Cal., in 1861; Perak, Malay Peninsula, Scortechini, Kunstler 1882, King's collector 1882. *A flamescens, Gungong, Pahang, 5,000 to 6,000 ft. Malay Peninsula, L. Wray and H. C. Robinson, 1905.

* The genera marked with an asterisk are not strictly Indian but they have been mentioned to indicate the type of Conifers extending down towards the Malay Peninsula and beyond.

Summary.

The wild conifers of the Indian Empire are confined to the Western and Eastern Himalayas. Hooker enumerates thirteen genera and twenty-five species of which twenty-three are wild. Of these again *Agathis loranthifolia* and *Dacrydium elatum* are wild in the Malay Peninsula. A list noting the actual places of occurrence from which the plants have been collected, as far as can be ascertained from the sheets of the Herbarium specimens of the Royal Botanic Garden, Calcutta, has been supplied. The collection of the conifers dates as early as 1812.

A short note has been added on the distribution of each of the individual wild Indian species of conifers as much as could be gathered from the data available.   But this statement requires further confirmation by explorations in the little known regions of Persia, Afghanistan, Baluchistan, Nepal, Sikkim, Bhootan, Tibet, Northern Burma and the most interesting spot from the standpoint of distribution of Floras— namely—the frontiers of Tibet, S. China, Yunnan, Szechuan, Northern Siam and Northern French Indo-China. But it may be remarked that generally speaking *Podocarpus neriifolia* and *Pinus Merkusii* have more

of Malayan element than Indian. *Juniperus communis* on the other hand forms perhaps an easterly link of the great belt of distribution of this species from Central and Southern Europe to Persia ending in the Western Himalaya. *Taxus baccata* has also a rather uniform range of long distribution from Europe through Persia to Himalaya ending in the Chino Tibetan border lands. The rest of the Indian species are indigenous to India, and are mainly confined to the Himalayan ranges in their wild state of growth forming frequently mixed associations with one or several species of Conifers.

List of Literature Consulted.

AITCHISON, J. E. T.—Catalogue of the plants of the Punjab and Sindh, London, 1869.

BENTHAM, G. and HOOKER, J. D.—Genera Plantarum, Vol. 3, pp. 420–442, London, 1847.

BLUME, C. L.—Rumphia, Vol. 3. t. 173, Lugdeneei, 1837.

BOISSIER, E.—Flora Orientalis, Vol. 5, pp, 705–711, Lugdeneei, 1881.

BOURDILLON, T. F.—The Forest Trees of Travancore, 1908.

BRANDIS, D.—Indian Trees, pp. 688–697, London, 1906.

BROWN, R.—A monograph of the Coniferous Thuja L. and of the North American species of the genus Libocedrus, 1847.

BURN-MURDOCH, A. H.—Trees and timbers of Malay Peninsula, Singapore, 1911.

BURKILL, I. H.—A working list of the flowering plants of Baluchistan pp. 86, Calcutta, 1909.

CLINTON-BAKER, H. and JACKSON, A. B.—Illustrations of Conifers, Vols. 1–3, Hertford, 1909–1913.

COLLET, C. S. H.—Flora Simlensis, Calcutta and Simla, 1902.

COOKE, T.—The Flora of the Presidency of Bombay, Vol. II, London, 1908.

COULTER, J. H. and CHAMBERLAIN, CH. J.—A morphology of Gymnosperms, Chicago, 1917.

COWAN, A. M. and COWAN, J. M.—The trees of Northern Bengal including shrubs, woody climbers, bamboos, palms and tree ferns, Calcutta, 1929.

DALLIMORE, W. and JACKSON, A. B.—A handbook of Coniferae including Ginkgoaceae, London, 1923.

DON, D.—Prodromus Florae Nepalensis, London, 1825.

ENDILCHER, S.—Synopsis Coniferarum, Sangalli, 1847.

ENGELMAN, G.—Revision of the genus Pinus ;—Trans. St. Louis. Acad. Soc. Vol. 4, St. Louis, 1830.

ENGLER, A.—Coniferae ;—Syllabus der Pflan. Fam., pp. 67–69, Berlin, 1898 and pp. 104–108, Berlin, 1919.

ENGLER, A. and GILG., E.—Gymnospermae—Die. Natürl. Pflan. Fam. Aufl. 2, Band 13, Leipzig, 1926.

FLORIN, R.—" Untersuchungen zur stammesgeschichte der Coniferales und Cordaitales", Erster-Tiel, Kungl. Sven. Vet. Handl. Tredje Serien, Bd. 10, No. 1, Stockholm, 1931.

FOXWORTHY, F. W.—Commercial timber trees of Malay Peninsula, Singapore, 1927.

FOXWORTHY, F. W.—Philippine Gymnosperms – Phil. Journ. Science. Bot. Vol. 4. No. 3, Manila, 1911.

FRITSCH, F. E.—The use of anatomical characters for systematic purposes, New. Phyt. Vol. 2, London, 1902.

GAMBLE, J. S.— A manual of Indian Timbers, London, 1901.

GHOSE, S. L.—A contribution of the morphology of Agathis Ovata (Moore) W. B. Journ. Ind. Bot. Soc. Vol. 4, 1924.

GRIFFITH, W.—Notulae ad Plantas Asiaticas (Posthumous papers) Pt., I–IV. Calcutta, 1847–54.

HARLOW, W. M.—The identification of the Pines of the United States, natives and introduced by needle structure, Bull. of the New York State College of Forestry, Vol. VI. No. 2a, 1931.

HEMSLEY, W. B.—The Flora of Tibet or High Asia, Jour. of the Linn. Soc. Bot. Vol. XXXV. London, pp. 124–265. 1902.

HENRY, A.—A new genus of Coniferae (Fokiena Henry et Thomas) Gard. Chro. Ser. 3., Vol. 49, London, 1911.

HOOKER, J. D.—Coniferae—Flora Brit. Ind. Vol. 5, London, 1890.

HOOKER, W. and J. D.—Icones Plantarum, Vol. 6. 1843, Vol. 11, 1867–71 and Vol. 19, London, 1889.

HOOKER, J. D., DARWIN, C. R., JACKSON, D.—Index Kewensis, Tom. 1–2. 1845 with suppl. 1–7, London, 1886–1925.

KANJILAL, U.—Forest Flora of the Siwalik and Jaunsar Forest Divisions of the United Provinces of Agra and Oudh, Calcutta, 1912.

KASHYAP, S. R.—Some aspects of the Alpine vegetation of the Himalaya and Tibet, (Presidential address) Botany of the 19th Indian Science Congress, 1932.

KIRKWOOD, J. E.—Forest distribution in the Northern Rocky mountain, Montana, 1922.

KURZ., S.—Forest Flora of Brit. Burma., Vol. 2, Calcutta, 1877.

MADDEN, E.—Observation on Himalayan Coniferae, etc. Journ. Agri-Hort. Soc. India, Vol. 7, Calcutta, 1850.

MARQUAND, C. V. B.—The botanical collections made by Captain Kingdon Ward in the Eastern Himalaya and Tibet in 1924–25, Jour. of the Linn. Soc. Bot. Vol. XLVIII, No. 321, London, 1929.

MASTERS, M. T.—A general view of the genus Cupressus—Journ. Lin. Soc. Bot. London. Vol. 31, 1895.

MASTERS, M. T.—Some features of interest in the order of Conifers, London, 1891.

MASTERS, M. T.—A general view of the genus Pinus—Journ. Lin. Soc. Bot. London, Vol. 35. 1901–1904.

MATSMURA. J.—Coniferae—Index. Plant. Japonicarum, Vol. 2. Tokyo, 1900.

MOLISCH, H.—Pflanzenphysiologie als Theori der Gartneri, Jena, 1916.

PARKER, R. N.—The cultivation of Conifers in Northern India—Indian Forester, Vol. 50 and 51. Allahabad, 1924 and 1925.

PARKINSON, C. E.—A forest flora of the Andaman Islands, Simla, 1923.

PARSONS, T. H.—An alphabetical list of Plants. Royal Bot. Garden, Peradeniya, Colombo, 1926.

PILGER, R.—Taxaceae—Engler, A.—Das. Pflanzenreich. IV, 5, Heft. 18, Leipzig, 1903.

RIDLEY, H. N.—The Flora of the Malay Peninsula, Vol. V, London, 1925.

ROXBURGH, W.—Flora indica—Clarke's edition, Calcutta, 1874.

SAHNI, B.—Revision of the Indian Fossil Plants, Pt. 1. Coniferales (a impressions and incrustations) Mem. G. S. India, New Series, Vol. XI, Calcutta, 1928.

SAXTON, W. T.—The classification of Conifers—New Phyt. Vol. XII, London, 1913.
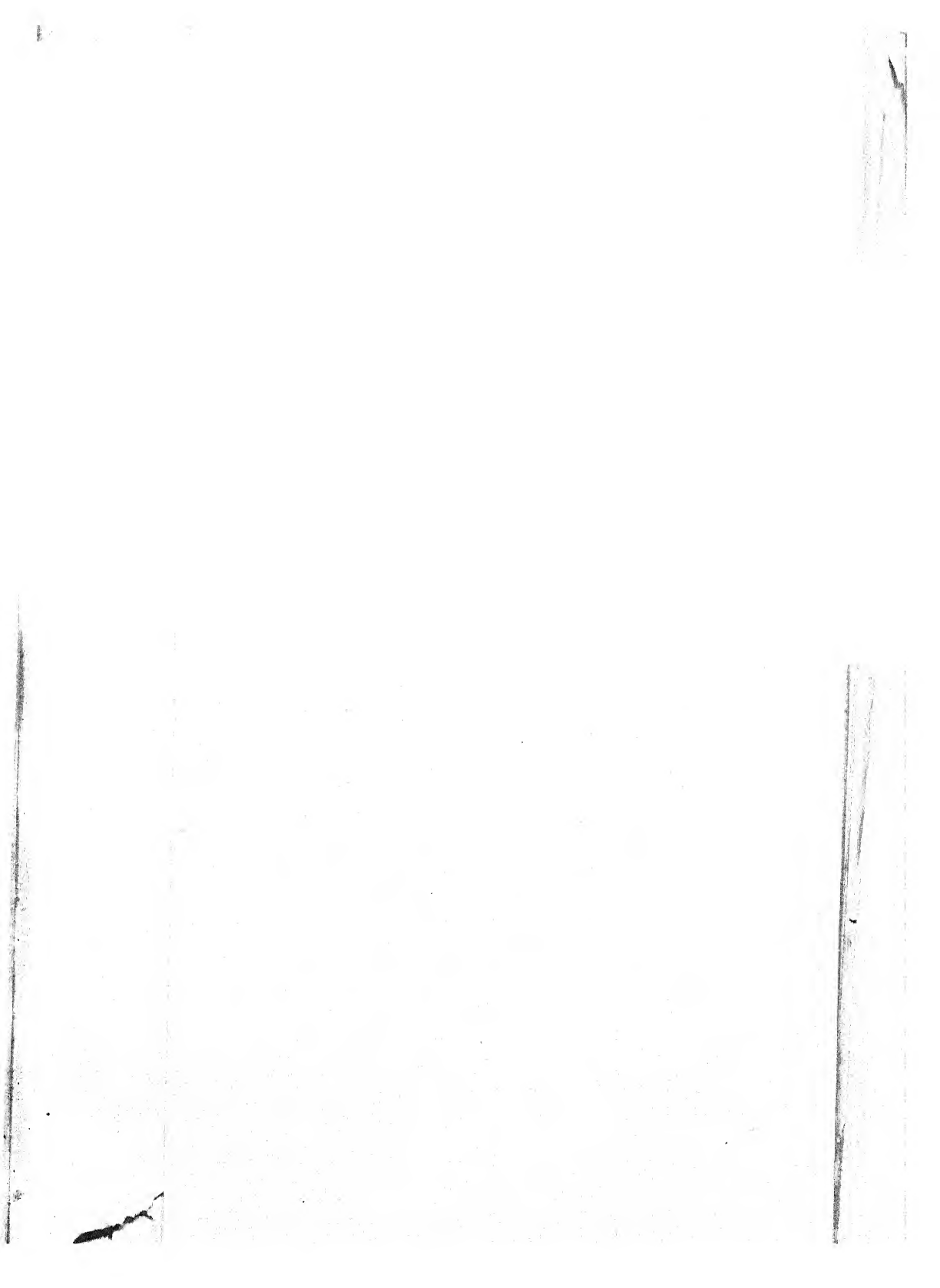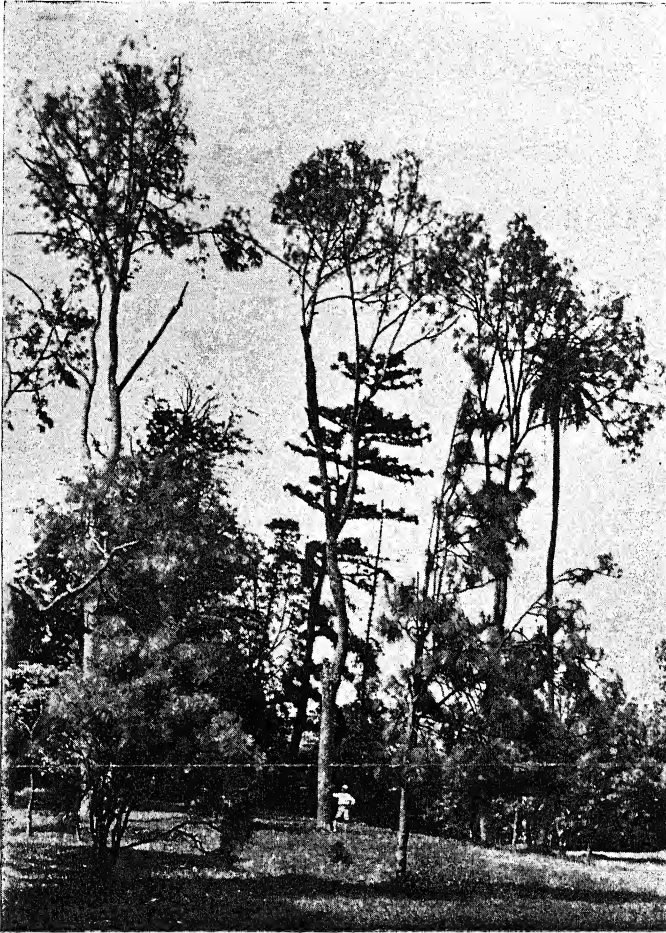
A group of Conifers cultivated in the Royal Botanic Garden, Calcutta *Pinus longifolia* in the fore-ground. *Araucaria cunninghamii, A. Cookii* and *A. Bidwillii* in the back-ground.

STAPF, O.—Index Londonensis, Tom. 1–6. London, 1929–1931.

TROUP, R. S.—The Silviculture of Indian Timber Trees, Vol. III, Oxford, 1921.

VEITCH, J.—A manual of Coniferae, Chelsea, S. W., 1881.

WALLICH, N.—Plantæ Asit. Rariores, Vols. 1–3, London, 1830–1832.

Explanations of Plates.

PLATE I.

[1] Map of the Indian Empire illustrating roughly the generic distribution of wild Conifers in the Indian Empire.

PLATE II.

Group of *Pinus longifolia* as cultivated in the Royal Botanic Garden, Calcutta. Some of the taller specimens are said to have been planted as early as 1794 by William Roxburgh, the then Superintendent of the Hon'ble East India Company's Garden by which name the Royal Botanic Garden, Calcutta, was known at that time.

HERBARIUM

ROYAL BOTANIC GARDEN, CALCUTTA,

14th October, 1931.

---

[1] Explanatory Note of the Map (Plate I).

In this map only the generic distribution of wild Conifers has roughly been shown. The predominance of the Conifers both in the number of different species and in the number of individuals is observed more in the Western Himalaya especially along Kumaon and Garhwal ranges of mountains extending up to Tibet and Central Nepal. To plot all the genera illustrating their specific distribution as well in this part near the Central Himalaya is a difficult task, as most of the species run along more or less in the same line varying more or less in elevations. The altitudinal variations as well as variations in the local distribution has been maintained as far as possible. Moreover, different species of one genus has different range of distribution as noticed in *Cupressus, Juniperus* and *Pinus*. Attempts have, therefore, been made to represent in a general way the distribution of these different species of each of the genera as much as can be managed in the space available.

A NOTE ON THE VARIATION OF LEAF IN RAILWAY CREEPER

BY

B. N. SINHA.

What we commonly know as Railway Creeper, is botanically the convolvulous member *Ipomaea pulchella*, Roth. (*I. palmata*, Forsk.) which is abundantly cultivated on railway platforms. It is a perennial twiner and bears purple, voilet or white flowers almost all the year round.

The leaves of this plant accommodate themselves in relation to each other rather very nicely and they form what is known as leaf-mosaic.

The leaves are palmate and are lobed almost to the very base. In the matter of lobation, we find a good deal of variation. A collection of such leaves was made at Indore in 1930, and the conclusions then arrived at, have recently been tested at Cuttack. The results obtained are concordant.

The commonest number of lobes in a leaf is five and it has, therefore, been regarded as the central or the mother-type. The higher number of lobation—the rarity of which increases with its increasing number—in a leaf, is certainly a dynamic modification of the central type, perhaps under cultivation.

The possible line of variational change is indicated in the text-figure where it is at once clear that No. 1 is the mother-type round which others are but simple variants.

The left lower lobe of No. 2 and right lower lobe of No. 3 and both the right and left lobes of No. 4, have already produced one lobule each. An indication—though an incipient one—of the formation of another lobule on the left lobe of No. 3 is, however, clearly discernible. Nevertheless, No. 4 condition becomes gradually very much more developed, as we follow Nos. 5–7. A step in advance, in the state of affairs, is found in Nos. 8–9 where the right lobule in its turn is found to be producing a further lobulation with the consequent result that in No. 9 we have a total of eight lobes.

A glance at the text-figure brings out a very striking feature namely that the two lowermost lateral lobes (of the mother-type) alone seem to have taken part in the modifications referred to and the three central ones have so to say remained static. It will then

further be seen that after these two laterals have produced their quota of one lobule each, they also like the central trio become inert and they, so to say, pass on the duty of any further lobulation to the two newly formed lobules.
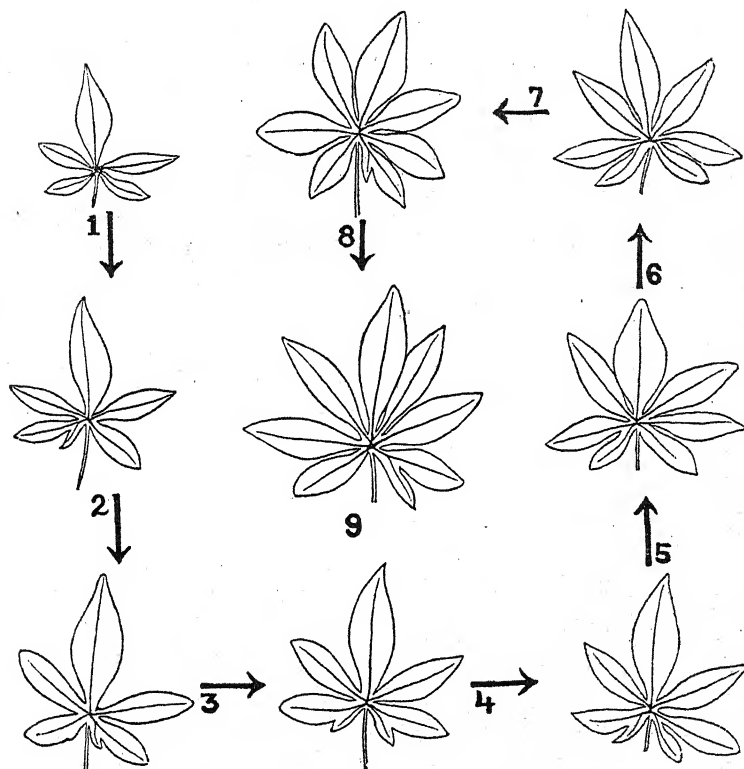
Text-Figure: *Ipomaea pulchella*, Roth. (Railway Creeper): Leaves showing variation of the lobes. The possible line of modification is indicated by the arrows. $\times \frac{1}{2}$.

The facts presented in this note are rather interesting from the point of view of variation but they are left as they are without entering into any discussion.

BOTANY DEPARTMENT,
RAVENSHAW COLLEGE,
CUTTACK.

ON SOME ABNORMAL LEAVES OF GINKGO

BY

B. SAHNI, SC.D.,

Professor of Botany, University of Lucknow.

While searching for flowers on a young maiden-hair tree at Lahore in 1920 I noticed a few abnormal leaves such as those shown in figure 1. The two lower margins of the triangular lamina, which in a normal leaf converge into the petiole, were bent over on the upper side and were more or less completely fused together so as to make a funnel. The same feature, with slight variations, was seen in 1924 on two trees at Mussoorie (West Himalaya) and more recently on one of the two plants at Cambridge (England) which are being trained as creepers against the south wall of the Botany School. In one case there were two little funnel-shaped pockets placed side by side at the base of the lamina (fig. 1 f). During the last twelve years I have searched in vain for similar leaves on many other trees, e.g., at Dehra-Dun, Calcutta, Vienna and the *Ginkgo* avenue in Dresden. Most of the abnormal leaves were gathered from the big male tree at the Municipal Gardens, Mussoorie, where a dozen were collected only from the lower branches; the other trees yielded only two or three specimens each.

The creepers at Cambridge were grafts from Montpellier, the plant which yielded the abnormal leaves being a female; the origin of the other trees is not known to me. The maiden-hair tree is such a familiar plant in the Far East that this abnormality will probably be well known in that part of the world, but I have not come across any reference to it in the literature. Professor Seward, who was kind enough to read this note in typescript, has recently informed me that he has observed this feature more than once.

Description.

As fig. 1 shows, all transitions are to be found between a normal flat lamina and a complete funnel. Even in the normal leaves the slightly thickened lower margins of the lamina (involute in the bud) often meet in an angle on the adaxial face of the petiole, while the more or less attenuated margins of the petiole may be continued some distance beyond this angle.[1] Velenovsky describes the same feature in a more marked degree in the leaves of seedlings.[2] In figs. 2-4 the

---

[1] Seward and Gowan (1900) pl. IX, figs. 39, 41; Sprecher (1907) figs. 57-59, 64, 65.

[2] Velenovsky (1907) p. 457, fig. 291 a, D-E.

structure of three funnel-leaves as seen in serial transverse sections is diagrammatically shown. Even in a normal leaf, the two petiolar bundles are somewhat inclined towards each other; in an abnormal
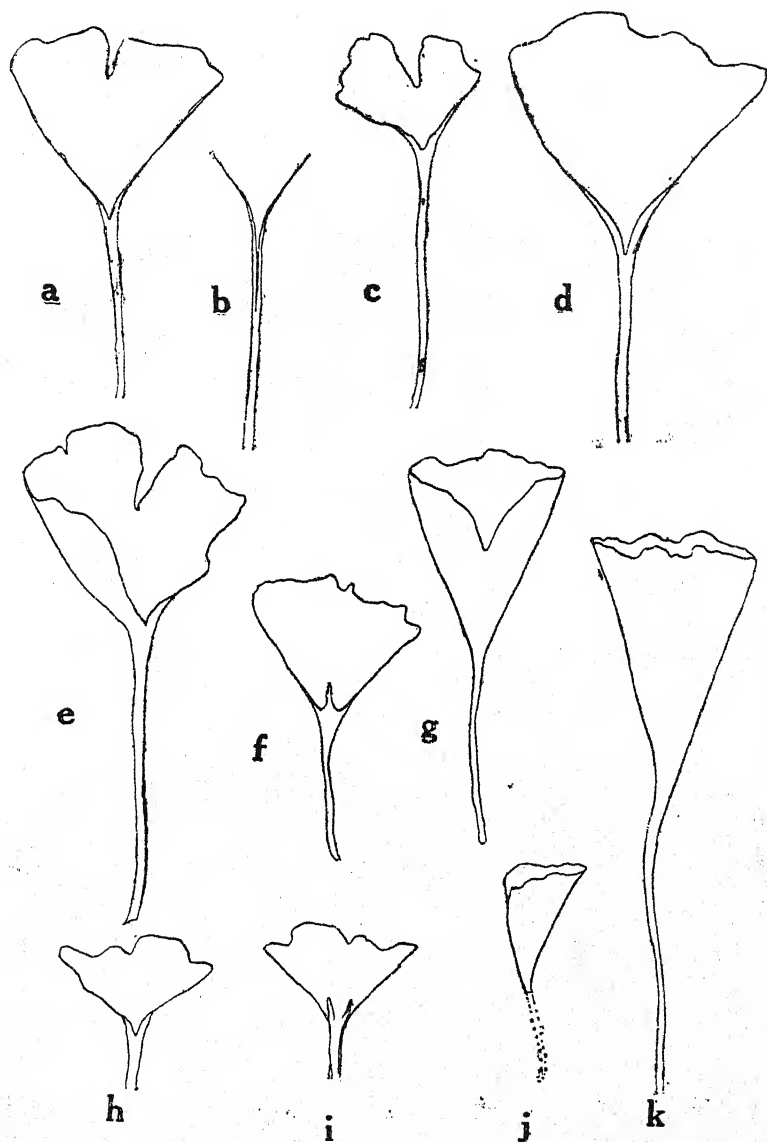
Fig. 1.

Fig. 1. Outline sketches showing transitions from the normal leaf (*a, b*) to fully formed ascidia (*k*). In *f* there are two conical pockets at the base of the lamina. In *i* (showing the dorsal surface of the leaf *h*) the petiole is continued into two horn-like processes adnate to the dorsal surface of the lamina. All × 1.

leaf they turn round and directly or almost directly face each other by their xylems, before they begin to undergo branching. The resulting bundles are placed in a ring with the xylem inwards. The central cavity begins to appear as a simple or branched slit lined by a cuticle ; the stomata are confined to the outer (morphologically abaxial) surface.

Fig. 4 is interesting as it shows a funnel within a funnel. In the microtome series (unfortunately incomplete)[1] there is no sign of a connexion between the two funnels ; but the inverted orientation of the bundles in the inner ring seems to leave no doubt that this is not a case of two separate leaves accidentally placed one within the other. The inner funnel evidently originated as a solid petiole (fig. 4 a) with the usual pair of bundles, but these are inclined towards each
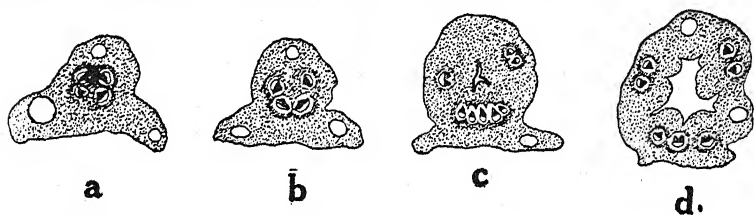
Fig. 2.
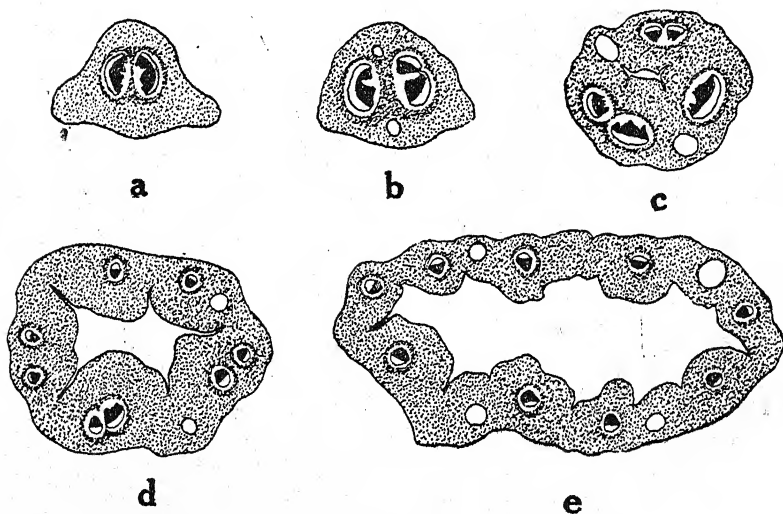
Fig. 3.

other by their phloems, not the xylems ; the stomata, too, as expected, are here confined to the concave side of the funnel. The whole structure thus follows the usual law of inversion.[2]

---

[1] The sections were made by a pupil, as an exercise in microtechnique.
[2] See Velenovsky (1907) p. 410 ff and literature there cited.

Discussion.

My object in writing this note is to describe rather than to attempt a theoretical interpretation of these abnormalities. But a few comparative remarks may be allowed. Ascidia are well known to occur both as a normal and an abnormal feature in many plants; [1] a similar form is often assumed by floral organs, especially stamens and petals, when modified as nectaries. Indeed the peltate leaves so often found in Angiosperms are not essentially different from ascidia. In certain garden varieties of *Codiaeum variegatum* (familiar in gardens under the wrong name of *Croton*) and in *Ficus Krishnae* [2] we have well known cases of leaves variously modified into ascidia.

It is probably futile to attach a morphological significance to the ascidia in *Ginkgo*. In some well known abnormalities described by Shirai, Fujii, Sprecher, Sakisaka and others [3] the collar of the ovule is replaced by a leaf-like lamina; in others a leaf is found bearing one or more ovules or stamens. The lamina in some of these abnormalities tends to envelope the base of the ovule like a cupule. No doubt some of the ascidia described in the present paper recall the cupules of some Pteridosperms. Externally at least, there is a considerable resemblance with *Whittleseya elegans* which Prof. Halle [4] has recently shown to be a campanulate spore-bearing organ. But this resemblance by itself may have no theoretical significance, especially as no ovules or microsporangia have been found enclosed in the ascidia here described.

I have said above that even in normal flat leaves the margins of the lamina often meet on the adaxial surface of the petiole, as in fig. 1 a. The formation of a pocket at the base of the lamina is thus only an exaggeration of the same feature. It is interesting to find that this feature has been figured in several Mesozoic and Tertiary leaves variously referred to the genera *Ginkgo*, *Ginkgoites*, *Ginkgodium* and *Baiera*: *Ginkgo lepida* Heer [5], *Ginkgoites antarctica* Sap. [6], *Ginkgodium Nathorsti* Yok. [7], *Ginkgodium gracile* Tateiwa [8], *Ginkgoites pluripartita* (Schimper) [9]. In reply to an enquiry Dr. T. M. Harris of Cambridge writes that he has also found it in some of his Greenland specimens of Rhætic Ginkgoales; and I

---

[1] For examples see the general works on plant morphology and teratology by Goebel, Velenovsky, Penzig, Masters, Worsdell, etc.

[2] Velenovsky (1907) p. 410. fig. 262; Molisch (1930) pl. II.

[3] Fujii (1896); Sprecher (1907) p. 144, fig. 161; Sakisaka (1929).

[4] Halle (1930) p. 472–73.

[5] Yokoyama (1906) pl. 9, fig. 2b.

[6] Saporta et Marion (1885) p. 142 fig. 71A; Sprecher (1907), p. 183, fig. 209.

[7] Yokoyama (1889); Seward (1919) p. 63, fig. 659A; Oishi (1931) p. 70.

[8] Oishi (1931) p. 70.

[9] See Seward (1926) pl. 9, fig. 86.

am indebted to him for photographs of a leaf of *Gingoites obovata*
Nath. which shows this peculiarity. Dr. Harris, too, holds the view
that this character is of diagnostic value. He has, in fact, already
used it to confirm his conclusions (based on cuticles) that one of his
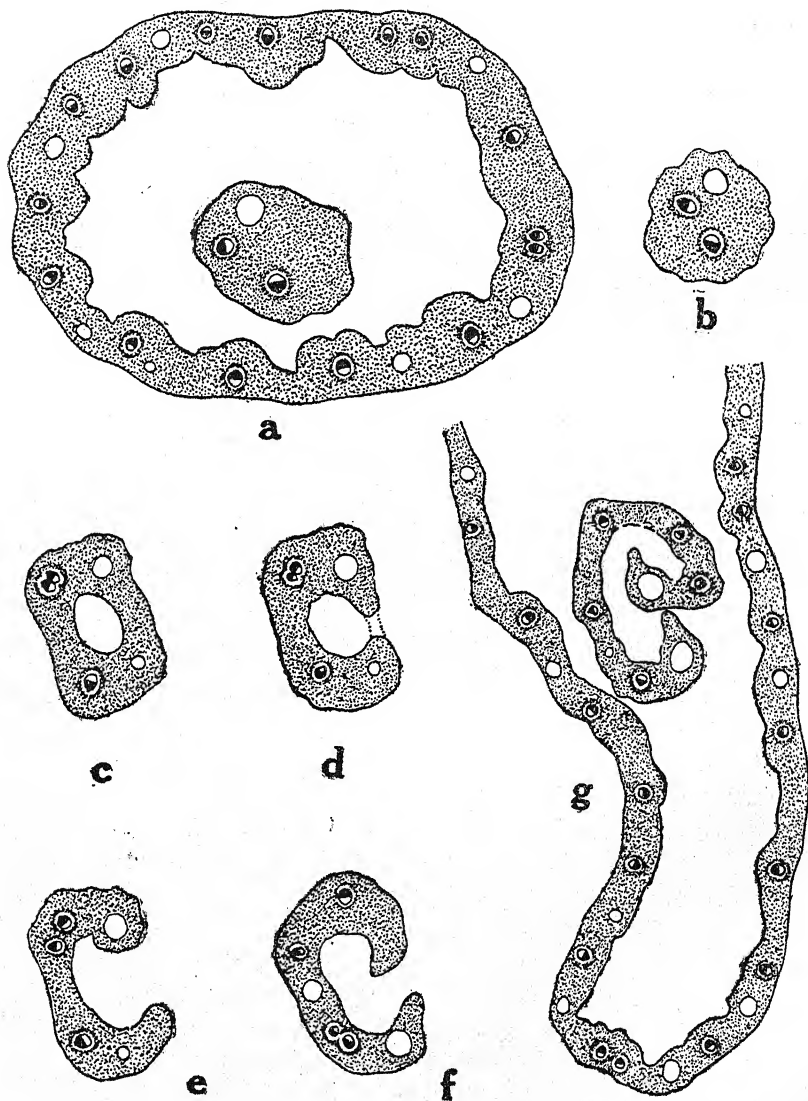

Fig. 4.

Figs. 2–4.   Microtome sections of three different ascidia. In figs. 2, 3 the
adaxial side is shown facing downwards.   In fig. 4 *b–f* the outer funnel is omitted.
Xylem black, phloem white. All × *ca.* 20.

Phoenicopsis-like leaves was, and that another was not, Ginkgoalean. Whether the same peculiarity existed in Palaeozoic members of the group it is difficult to say. In fact the attribution of many of the older leaves, such as species of *Psygmophyllum*, *Rhipidopsis* and other genera, to the Ginkgoales is still open to doubt. But if this character is found among any of these Palaeozoic forms it would strongly support their reference to that group. In an interesting paper recently published Dr. O. Posthumus [1] has shown that certain fossil fern leaves (*Dictyophyllum*, *Camptopteris*) resemble those of some living Dipteridineae in a peculiar twist in the base of the lamina : a welcome corroboration of their dipterid affinities, already suspected on other grounds.

It is indeed strange how these little peculiarities sometimes tend to persist through geological time.

Bibliography.

FUJII, K. (1896)—On the different views hitherto proposed regarding the morphology of the flowers of *Ginkgo*. Bot. Mag. Tokyo. Vol. X.

HALLE, T. G. (1930)—Fifth International Botanical Congress (Cambridge, 1930) Report of Proceedings (Section of Palaeobotany).

MOLISCH, H. (1930)—Biologische Forschungen in Indien. Vortr. d. Ver. Verbr. nat. Kennt. in Wien, 70 Jahrg.

OISHI, S. (1931)—Mesozoic Plants (in Japanese) p. 70.

POSTHUMUS, O. (1928)—Einige Eigentümlichkeiten der Blattform bei *Dipteris* und bei anderen noch lebenden oder fossilen Pflanzen. Rec. trav. bot. néerl. XXV.

SAKISAKA, M. (1929)—On the seed-bearing leaves of *Ginkgo*. Jap. Journ. Bot. 4.

SAPORTA ET MARION (1885)—Lévolution du regne végétal. Tome 1.

SEWARD, A. C. and GOWAN (1900)—The maiden hair tree (*Ginkgo biloba*). Annals of Botany, Vol. XIV.

SEWARD, A. C. (1919)—Fossil Plants Vol. IV.

SEWARD, A. C. (1926)—The Cretaceous plant-bearing rocks of East Greenland. Phil. Trans. Roy. Soc. Vol. 215.

SPRECHER, A. (1907)—Le *Ginkgo biloba*.

VELENOVSKY, J. (1907)—Vergleichende Morphologie der Pflanzen, vol. II.

YOKOYAMA, M. (1889)—Jurassic plants from Kaga, Hida and Echizen. Jour. Coll. Sci. Imp. Univ. Tokyo. Vol. III.

YOKOYAMA, M. (1906)—Mesozoic plants from China. Journ. Coll. of Sci. Imp. Univ. Tokyo. Vol. XXI.

---

[1] Posthumus (1928).

CHROMOSOME NUMBERS OF SOME SOLANACEOUS PLANTS OF BENGAL

BY

PARAM NATH BHADURI, M.SC.,

Department of Botany, Calcutta University.

Vilmorin and Simonet (14) in a recent paper have given a comprehensive account of the chromosome numbers of species belonging to the family Solanaceae. Their investigation deals mainly with the European species and adds considerably to our knowledge of the chromosome number of Solanaceous plants. As nothing was known of the chromosome complements of the Solanaceous plants of Bengal, the present investigation was undertaken with the idea of adding some more data to that collected by workers abroad, and incidentally re-examining the question of polyploidy in the genus Solanum, in the light of the results obtained. The chromosome number of some of the species which had already been determined by other workers, were re-investigated once again, as it was thought that the cytological history of the plants might not be the same.

Material and Methods.

The material used in this investigation was collected from different localities round about Calcutta. The material was fixed on bright days between 11 a.m. and 3 p.m., in the field. To facilitate penetration of the fixing fluid the calyx and the top of the corolla were removed from the flower buds, leaving the base of the corolla as a ring round the ovary, bearing the epipetalous stamens. They were first dipped in Acetic-alcohol (1 : 2), to remove the waxy coating from the stamens, and then fixed in Allen's modified Bouin's fluid. The material was then dehydrated, cleared and embedded in the usual way. Sections were cut 8 to 10 μ thick and stained with Haidenhain's iron alum haematoxylin.

Belling's iron aceto-carmine method was also employed to obtain conformatory results with fresh material. This method worked satisfactorily with all the species.

Observations.

The chromosome numbers of the species investigated were chiefly computed from the meiotic stages of the microspore mother cells. Check counts were taken from flowerbuds of the same, and different plants, to eliminate any possible source of error.

It will be noted from the above table that the different species of Solanum have 12 as the haploid chromosome number. *Solanum nigrum* L., however, gave very interesting results, and showed distinct polyploidy within the species. Jorgenson and Crane (9) who corroborated Winkler's (15) observations have shown that *Solanum nigrum* L. has 36 haploid chromosomes. They also found that *S. nigrum* var. *Gracile* Raddi, which resembles *S. nigrum* L. in all morphological characters
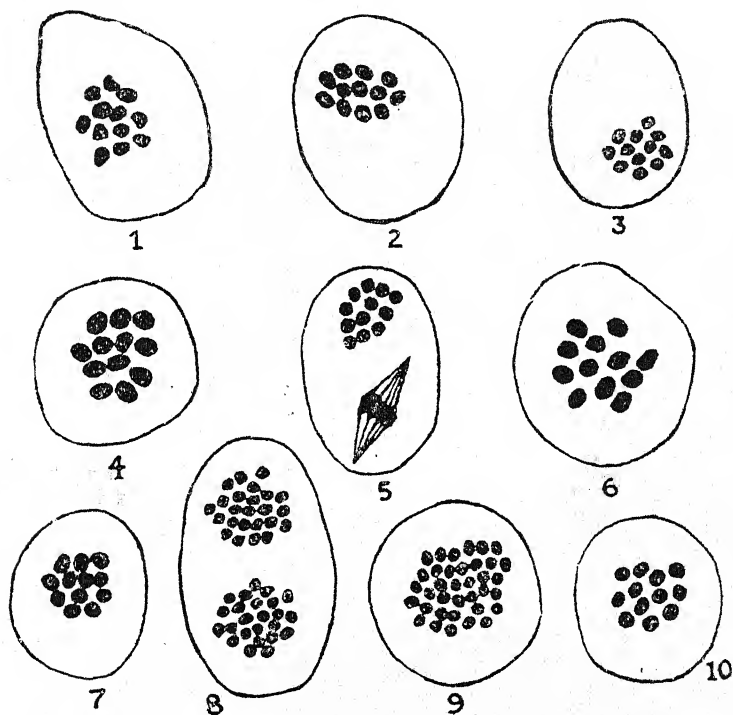
Text-figure I. Figs. 1–9, Solanum. 10. Lycopersicum.

1. *Solanum indicum* L. Heterotypic metaphase. 2. *Solanum xanthocarpum* Schrad & Wendl. Heterotypic metaphase. 3. *Solanum torvum* Swartz. Heterotypic anaphase polar view. 4. *Solanum micranthum* Wild. Heterotypic metaphase. 5. *Solanum verbescifolium* L. Homotypic metaphase. 6. *Solanum trilobatum* L. Heterotypic metaphase. 7. *Solanum nigrum* L. (2n). Heterotypic metaphase. 8. *Solanum nigrum* L. (4n). Homotypic metaphase. 9. *Solanum nigrum* L. (6n). Heterotypic metaphase. 10. *Lycopersicum esculentum* Mill. Heterotypic metaphase. × 1,100.

excepting in their more slender habit and more even outline of the leaves has also 36 haploid chromosomes. Vilmorin and Simonet (14) found a plant, identical with *S. gracile* Otto, (equivalent to *S. gracile* Link), which resembles *S. nigrum* L. in all morphological characters but possesses 12 haploid chromosomes.

Critical observations of *S. nigrum* L. for over two seasons have brought to light the hitherto unnoticed fact that plants identified as *S. nigrum* L. by Prain (12), Hooker (8) and others, differ considerably in both morphological and cytological characters. The important morphological characters of the three types of *S. nigrum* L. with their chromosome numbers, and also the morphological characters of *S. nigrum* L. (n = 36) as given by Jorgensen and Crane (9) are given below in Table II.
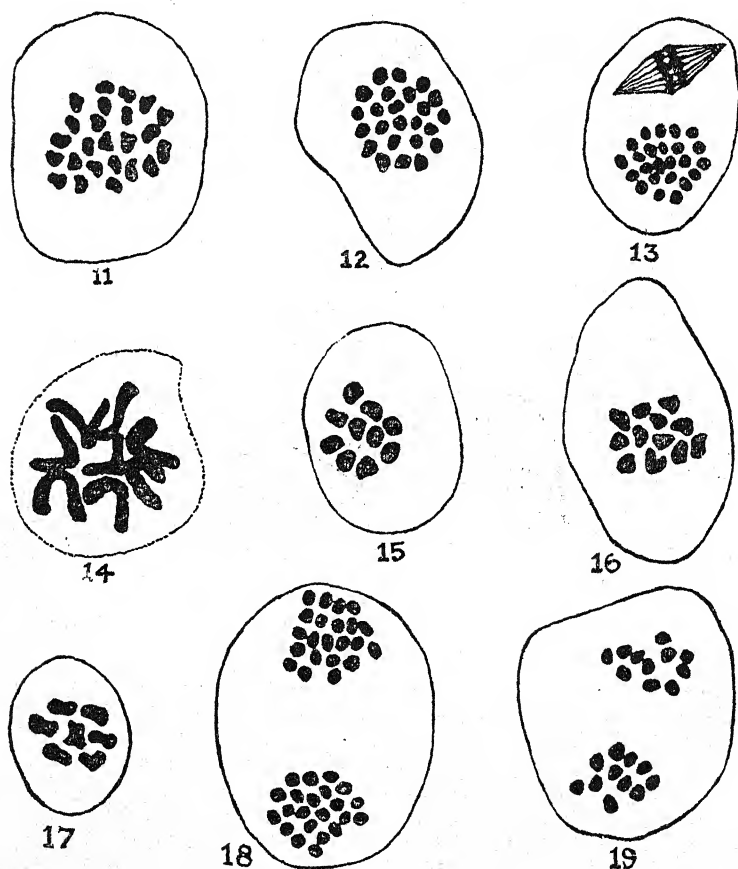
Text-figure II. Figs. **11.** *Physalis peruviana* L. Heterotypic metaphase. **12.** *Physalis minima* L. Heterotypic metaphase. **13.** *Withania somnifera* Dun. Homotypic metaphase. **14.** *Cestrum nocturnum* L. Mitotic division in the embryo sac. Metaphase, polar view. **15.** *Nicotianaplumbaginifollia* Viv. Heterotypic metaphase. **16.** *Datura fastuosa* L. Heterotypic metaphase. **17.** *Petunia nyctagi-niflora* Juss. Heterotypic metaphase. **18.** *Salpiglossis sinuata* Ruiz. Homotypic metaphase. **19.** *Brunfelsia americana* Sw. Homotypic metaphase. × 1,100.

The following table gives an account of the plants that had been worked out, their chromosome numbers, and the names of the investigators.

Table I.
Chromosome Numbers in Solanaceae.

Name of the plant.	Haploid chromosome number	Investigator.
Solanum xanthocarpum Schrad and Wendl.	12	Jorgensen (10) Vilmorin and Simonet (14) and Present writer.
„ *indicum* L.	12	Present writer.
„ *verbascifolium* L.	12	„
„ *torvum* Swartz.	12	„
„ *trilobatum* L.	12	„
„ *micranthum* Willd.	12	Jorgensen (10) and Present writer.
„ *nigrum* L.	12	Present writer.
„ „	24	„
„ „	36	Winkler (15), Jorgensen and Crane (9), Vilmorin and Simonet (14) and Present writer.
Lycopersicum esculentum Mill.	12	Winkler (15). Lesley (11), Jorgensen and Crane (9), Vilmorin and Simonet (14), Cooper (4) and Present writer.
Physalis peruviana L.	24	Vilmorin and Simonet (14) and Present writer.
„ *minima* L.	24	Present writer.
Withania somnifera Dun.	24	Present writer.
Datura fastuosa L.	12	Belling and Blakeslee (1), Vilmorin and Simonet (14) and Present writer.
Cestrum nocturnum L.	Ca 8	Present writer.
Nicotiana plumbaginifollia Viv.	10	Christoff (3) and Present writer.
Petunia nyctaginiflora Juss.	7	Ferguson (6), Derman (5) and Present writer.
Salpiglossis sinuata Ruiz.	22	Vilmorin and Simonet (14) and Present writer.
Brunfelsia americana Sw.	11	Present writer.

TABLE II.

Morphology of Polyploid forms of *S. nigrum* L.

	DIPLOID *S. nigrum* L.	TETRAPLOID *S. nigrum* L.	HEXAPLOID *S. nigrum* L.	*S. nigrum* L. AS DESCRIBED BY JORGENSEN AND CRANE.
Habit and Branching.	Slender habit, internodes more elongated. Main stem very short, lateral branches given out almost from the ground level.	Main stem short and divides into two branches from a little height giving the appearance of pseudo-dichotomous branching.	Varying habit, either like diploid or tetraploid plants.	Branching almost pseudo-dichotomous and numerous small shoots develop from the axils of the leaves.
Nature and arrangement of leaves.	Spirally arranged, pseudo-opposite on the flowering stems. Petioled, ovate, Margin slightly dentate.	Spirally arranged, pseudo-opposite on the flowering stems. Ovate, petiolated. Margin markedly dentate.	Spirally arranged, pseudo-opposite on the flowering stems. Ovate petiolated. Margin slightly dentate.	Spirally arranged, pseudo-opposite on the flowering stems. Ovate. Margin usually dentate.
Infloresence ...	4-6 flowers. Peduncles 6-8 mm. Pedicels 3-4 mm.	5-7 flowers. Peduncles 8-10 mm. Pedicels 5-6 mm.	4-6 flowers. Peduncles 8-10 mm. Pedicels 5-6 mm.	5-11 flowers.
Flowers ...	7-9 mm. diameter.	11·5 mm. diameter.	11·5 mm. diameter,	···

	DIPLOID *S. nigrum* L.	TETRAPLOID *S. nigrum* L.	HEXAPLOID *S. nigrum* L.	*S. nigrum* L. AS DESCRIBED BY JORGENSEN AND CRANE.
Petals ...	Slightly ligulate, almost free, united at the base. Yellow spot at the base. 3–4 mm. long.	Triangular, fused to about half their length. No yellow spot at the base and not ligulate. 5 mm long.	Slightly ligulate almost free, fused near the base. 5–6 mm. long. Inconspicuous yellow spot at the base.	Ligulate 3–4 mm. long. White with a yellow spot at the base. Almost free.
Sepals ...	Short and obtuse. 3–4 mm.	Short and obtuse. 3–4 mm.	Short and obtuse. 3–4 mm.	Short and obtuse.
Stamens	Filament and anther of unequal lengths. 0·5 mm. + 1·5 mm.	Filament and anther of equal lengths. 1·5 + 1·5 mm.	Filament and anther of unequal lengths. 2 + 1 mm.	Free, short filament.
Fruit ...	Globose, shining blue-black, 4–6 mm. diameter.	Globose, orange-red. 5–6 mm, diameter.	Globose, dull-purplish black 6–8 mm. diameter.	Globose, shiny and bluish black, 5–8 mm. diameter.
Stem and hairs ...	Green with few ribs. Hairs very few, on stem and under surface of leaves and veins.	Green with purplish tint, prominent ribs. Hairs present, specially on younger parts, stems and veins.	Green with purplish tint, occasional ribs,	Few scattered hairs. Mostly occur on stems and veins of leaves.
Measurement of Pollen (average of 100 grains).	23.5μ	28.5μ.	29.5μ.	......
Haploid chromosome number.	n = 12	n = 24.	n = 36	n = 36

It will be seen from the above table that though the diploid and hexaploid plants resemble each other somewhat closely, yet in the latter the floral parts are markedly larger than in the former. Besides, the berry colour is very characteristic of the three types, and the plants can be discriminated readily in the field by this character alone. The tetraploid plants differ greatly from the other two types and has morphological characters approaching very near to *S. luteum* Mill., (= *S. tomentosum Lam)* as described by Jorgensen and Crane (9), from which it differs, however, in the size of the sepals and by the absence of the yellow spot at the base of the petals. As the diploid type of *S. nigrum L.* (n = 12) does not resemble (in all external features) *S. nigrum L.* (n = 36) of the previous investigators it is difficult to say whether the diploid type represents *S. gracile* Otto (n = 12), which according to Vilmorin and Simonet (14) closely resembles *S. nigrum L.* (n = 36) in all external characters.

It has been pointed out by several investigators that the polyploids differ from each other in both morphological and cytological characters. From Table II it will be seen, however, that though there is some difference in morphological characters between the polyploids, no gigantism of vegetative organs or cells have been noted. Gershoy (7) in Viola has shown that with each higher number of chromosome in the polyploids, the chromosome size decreases, while the volume of the nucleus and the pollen grain increases. Blakeslee and Belling have also found that increase in volume of pollen grains is associated with increase in chromosome number in the polyploid mutations of *Datura.* Measurement of pollen grains and pollen mother cells in the polyploids of *S. nigrum L.* however, failed to reveal any relationship with chromosome numbers and dimension of pollen grains.

It has also been pointed out by several investigators that the tetraploids are less fertile than the diploids. According to Sansome (13), in Tomato 75 per cent of the pollen grains in tetraploid plants are fertile as compared to 100 per cent. in the diploids, and a fruit of tetraploid produces, on average, only 20 seeds as compared with 90, per fruit, of a diploid. In the tetraploid and hexaploid plants of *S. nigrum L.* a sterility of 10 to 16 per cent in the pollen grains have been observed. The polyploids all have been found to fruit equally vigorously under favourable conditions.

In the case of the other plants investigated, the determination of chromosome numbers of the previous investigators have been mostly confirmed. Of the plants whose chromosome numbers have been determined for the first time *Physalis minima,* was found to contain 24 haploid chromosomes as in *Physalis peruviana. Brunfelsia ameri-*

cana, showed 11 haploid chromosomes. Campin (2) appears to be the only investigator who has found this number in Solanaceæ, but he is not quite definite. Considering the fact that both *Brunfelsia* and *Salpiglossis* belong to the tribe Salpiglossoideæ the chromosome numbers 11 and 22 do not appear to be at all surprising. The chromosomes in *Cestrum* appear to be very irregular in their shape and as such it was rather difficult to obtain definite evidence as to their number from the meiotic divisions of the microspore mother cells, and the counts were made from nuclei undergoing mitotic divisions in the embryo-sac.

Summary.

The chromosome numbers of some of the Solanaceous plants commonly occurring in Bengal have been determined. The chromosome numbers of the following plants have been determined for the first time :—

> *Solanum indicum* L. (n = 12)
> *Solanum verbascifolium* L. (n = 12)
> *Solanum trilobatum* L. (n = 12)
> *Solanum torvum Swartz* (n = 12)
> *Withania somnifera* Dun (n = 24)
> *Physalis minima* L. (n = 24)
> *Cestrum nocturnum* L. (n = Ca 8)
> *Brunfelsia americana* I Sw. (n = 11)

Polyploidy within the species have been noted in *Solanum nigrum* L. (n = 12, n = 24 and n = 36). The morphological characters of the polyploids have been given. The close resemblance of the tetraploid form of *S. nigrum* with *S. luteum* Mill. has been indicated.

Literature Cited.

1. BELLING, J. and BLAKESLEE, A. F. 1923.—The reduction division in haploid, diploid, triploid and tetraploid Daturas.   Proc. Nat. Acad. Sci. 9 : 106–111.

2. CAMPIN, M. G. 1924.—An irregular method of pollen formation in Solandra Grandiflora Sw. New. Phy. 23: 282–287.

3. CHRISTOFF, M. 1928.—Cytological studies in the genus Nicotiana. Genetics. 11: 267–279.

4. COOPER, D. C. 1931.—Macrosporogenesis and the development of the macrogametophyte of Lycopersicon esculentum.   Amer. Jour. Bot. 18 : 739–748.

5. DERMAN, H. 1931.—Polyploidy in Petunia.   Amer. Jour. Bot. 18 : 250–260.

6. FERGUSON, M. C. 1927.—A cytological and a genetical study in Petunia.   I. Bull. Torr Bot. Club. 64 : 657–664.

7. GERSHOY, A. 1928.—Studies in North American Violets. I. General considerations.   Vermont. Agric. Exp. Sta. Bull. 279 : 1–18.

8. HOOKER, J. D. 1875.—The Flora of British India, 4 : 229.

9. JORGENSEN, C. A. and CRANE, M. B. 1927.—Formation and morphology of Solanum chimeras.   Jour. Genet. 18 : 247–273.

10. JORGENSEN, C. A. 1928.—The Experimental Formation of heteroploid plants in the genus Solanum. Jour. Genet. 19 : 133–210.

11. LESLEY, M. M. 1926.—Maturation in diploid and triploid tomatoes.   Genetics. 2 : 267–279.

12. PRAIN, D. 1903.—Bengal Plants. 2 : 744–745.

13. SANSOME, F. W. 1929.—Polyploidy in the Tomato.   Conference on polyploidy held at John Innes Horticultural Institution.

14. ROGER VILMORIN de et MARC SIMONET. 1928.—Recherches sur le Nombre des chromosomes chez les Solanees. Zeitschr. f. ind. Abastam. Suppl. Bd. II. 1520–1536.

15. WINKLER, H. 1910.—Uber die Nachkommenschaft der Solanum Pfropfbastarde und die chromosomen zahlen ihrer keimzellen.   Zeitschr. f. Bot. 2 : 1–38.

THE SCENT AND COLOUR OF FLOWERS IN RELATION TO BIRD-POLLINATION

BY

T. C. N. SINGH.

The recognition of the importance of scent in relation to bird-pollination has been neglected in such a measure that in recent years the opinion has been expressed that birds make no use of their sense of smell and so none of the flowers habitually fertilized by them are fragrant [1]. During the course of his observations on pollination by birds in Indian flowering plants, the author has come across certain interesting facts with regard to scent and colour which call for some critical remarks.

The flowers of the Rubiaceous tree *Morinda tinctoria* Roxb. are cream coloured and when in bloom (*April* to middle of *June*), these emit a very strong pleasing sweet fragrance which spreads in a radius of at least one furlong. It is interesting to record that the mature flower is about one-fourth full of a sugary juice which is secreted by the nectary at the base of the corolla-tube. In the cool hours of morning between five and eight o'clock, this tree is visited almost exclusively by sun-birds (*Cinnyris asiaticus*), both male (steel-black: this is the colour which it acquires during the mating season) and female. The sugary juice is heartily feasted upon by these birds which in Indian vernacular are called *shakar-khorā* (or the sugar-eaters). This name may very fitly be extended even to other such birds. However, in this act of drinking the sugary juice from flower to flower, the sun-birds unconsciously effect pollination [2].

Thus we see that the flowers of *Morinda tinctoria* Roxb. are not only not scarlet but are at the same time sweet scented and are pollinated by sun-birds. The absence of scarlet colour and presence of scent and yet the occurrence of pollination by birds in a single species does really appear to be antagonistic to the prevailing idea of ornithophily. Scarlet colour, as is well known, is supposed to

---

[1] Hampton: *The scent of flowers and leaves*, 1925.

[2] The fuller paper, on the pollination of *Morinda tinctoria* Roxb. by sun-birds, will subsequently be published,

be common in ornithophilous flowers e.g. *Erythrina indica* Lam.[1], *caffra* Thunb.[2], *Christa-galli* Linn., *herbecea* Linn. and *speciosa* Andr.[3], several species of *Lobelia*, *Begonia fuchsioides* Hook., *Amherstia nobilis* Wall. and *Brownea coccinea* Jacq.[4], *Bombax malabaricum* DC. and several others; but on the other hand quite a number of plants e.g. *Butea frondosa* Roxb., *Euphorbia pulcherrima* Willd., *E. splendens* Boj. ex Hook., *Hibiscus rosa-sinensis* Linn, varieties of *Canna indica* Linn., American species of *Aechmea* and *Vriesea*[5], Malay Zingiberaceae[6] and many more may be enumerated, in which the scarlet colour of their bracts or corolla, does not exert any influence in attracting birds to perform the function of pollination; while there are others e.g. *Feijoa Schenckiana* Kiaersk, with snow-white flowers, *Marcgravia umbellata* Linn. with dull brown flowers, *Courouptia guianensis* Aubl. and *Weigela* sp. with deep carmine flowers and *Strelitzia Reginae* Banks with bright orange perianth and a large azure labellum[7], possessed of flower-colours, other than scarlet, which are regularly pollinated by birds.

So it is clear that we should relinquish the view, which of late has been gaining ground, of seeing the scarlet coloured flowers with an eye of suspicion and regarding most of them to be ornithophilous[8]. It is, therefore, proposed to examine and discuss in brief the characters which appear to be of most importance with regard to bird pollination.

The tree *(Morinda)* in bloom is leafy and is not in the least so conspicuous as the leafless *Bombax malabaricum* DC., *Erythrina indica*, Lam. and several other ornithophilous members with their scarlet flowers. In the latter, certainly colour does act as a guide which purpose in *Morinda tinctoria* Roxb. is presumably effected through the agency of the sweet fragrance of its flowers. And the author has also himself seen in the early hours of morning, sun-birds flying to this tree from distant places.

Already several investigators[9] have recognised and the fact is now fairly well established that scent plays an important rôle in the courtship of moths, butterflies, beetles and fruit-flies. Likewise, in *Morinda*, besides serving as guide, the scent perhaps plays a similar

---

[1] Singh: *Jour. Bomb. Nat. Hist. Soc.* XXXIII, 1929.

[2] Galpin: *Gard. Chronicle*, IX, 1819; Knuth: *Hand-Book of Flower Pollination*, I, 1908.

[3-4] Kerner: *The Natural History of Plants*, II, 1904.

[5-7] Schimper: *Plant Geography*, 1904.

[8] Hampton: (1925) *ibid.*

[9] Carpenter: *Proc. Eng. Entom. Soc.* 1914; Hampton: *loc. cit.*; Longstaffe: *Butterfly Hunting in Many Lands*, 1912; Knuth: *loc. cit.*

part in the way of fulfilment of the most important biological end-
the courtship-of the sun-birds. Because, these during the period of
the year intervening between April and June, are, so to say, at the
height of their mating fervour. This is evidenced by their sweet but
short pretty chirpings which they give out as they hop from bough to
bough and also by the steel-black colour of the males.

From a rather rapid survey related in the preceding paragraphs,
the following important details are at once apparent in the case of
certain Indian types :—

Name of Plant	Flower Colour	Scent	Nectary	Name of Bird
Morinda tinctoria Roxb.	Cream White	Strong pleasing fragrance.	Sugary juice produced in great abundance.	*Cinnyris asiaticus.*
Erythrina indica Lam.	Scarlet	None	Ditto	*Acridotheres tristis tristis*
Bombax malabaricum DC.	Scarlet	None	Ditto	Several birds.

Although the species briefly described here are far removed from
each other in the natural scheme of classification, yet the most impor-
tant fact that stands out prominently is the character of the sugary
juice which is secreted in great abundance by the nectary apparatus in
each case. This condition is not only characteristic of the species
referred to but it is so also of other ornithophilous species e.g.
Marcgravia [1], *Manettia* [2], *Protea* [3], *Erythrina caffra* Thunb. and
Tecoma capensis Linde. [4], *Ravenala madagascariensis* Gmel. and *Stre-
litzia Reginæ* Banks [5] and several others [6].

Thus, from a perusal of facts presented, it is clear that the relation
between bird-pollination and scarlet colour of a flower cannot be
upheld in the same terms as is often stressed and in the light of the

---

[1] Knuth: *loc. cit.* Vol. I ; Willis: *A Dictionary of Flowering Plants and Ferns*, 1919.

[2] Knuth: *loc. cit.* Vol. II : Müller: *The Fertilization of Flowers*, 1883 : Rendle: *The Classification of Flowering Plants*, Vol. II, 1925; Schimper ; *Plant Geography*, 1925.

[3] Schimper: *loc. cit.* ; [4] Galpin: *loc. cit,*; Knuth: *loc. cit.*

[5] Schimper: *loc. cit.* ; [6] Knuth: *loc. cit,*; Scott-Elliot: *Ann. Bot.* IV and V, 1889–91 ; Thomson: *Transc. & Proc. N. Z. Institute*, VIII, 1880 ; Kerner : *The Natural History of Plants*, II, 1904.

observations on pollination in *Morinda tinctoria* Roxb. the view with regard to scent (or fragrance), like that of colour, will also need have to be modified. In fact, these characters-scent and colour-which when present appear to be of secondary importance and help chiefly as guides to the pollinators. But, as shown above, the character of most importance and constant occurrence in relation to ornithophily, is the production of sugary juice in great profusion. This (sugary juice) by itself when stored up in petals (e.g. in *Feijoa Schenckiana* Kiaersk.) themselves or in some sort of a receptacle (due allowance, however, being made of the size of the bird and flower) formed by the close fitting or otherwise of the sepals or petals or both (as found in known ornithophilous flowering plants), is almost sure to attract birds as pollinators. The birds are, after all, gluttons and so they are naturally lured by those plants which offer them plenty of food rather than flaring colour or pleasing scent.

RAVENSHAW COLLEGE,
    CUTTACK.

It is with the deepest regret that we have to record the sudden and sad death of Dr. Winfield Dudgeon. The news came to us as a great shock. His connection with the Indian Botanical Society is well-known. He was not only its first President but also its founder. We have lost in him not only an able botanist but a kind and generous friend. We offer our most heartfelt sympathy to Mrs. Dudgeon in her bereavement.

<div style="border:1px solid black">

Obituary

Dr. WINFIELD DUDGEON.

1886—1932

</div>

Dr. Winfield Dudgeon, the first President of the Indian Botanical Society, Head of the Department of Biology in The Ewing Christian College, Allahabad, and part-time teacher in the University of Allahabad died in America on December 26, 1932. His premature death is deeply mourned by all of us, and to those who like the present writer, were more intimately associated with him, it is a great personal loss.

I counted it a privilege to be his student, and in this inadequate sketch of his life I have quoted freely from some of his letters, so that the readers may have an opportunity of gaining more intimate knowledge about his life and work.

I first came to know him in 1921, as a professor of Botany in the Ewing Christian College, Allahabad. After my Intermediate I began to come into closer contact with him. I felt so interested in his lectures that after passing my M.Sc. in Botany in 1927 I refused to do anything else and took up research in Angiosperm Morphology under his directions. I continued this for three years at Allahabad and even after my appointment at Agra in 1930 I kept in close correspondence with him and greatly profited by his advice and encouragement. I saw him last on April 28th, 1932, when he came to Agra on his way back from Aligarh to Allahabad only to see me before his departure to America on furlough for one year. But for him I should not have been a teacher of Botany, but should have been somewhere in the Agricultural Department of the Jaipur State.

He was an excellent teacher. While he was not very eloquent, his keenness in the subject gave a reality to it. To the Intermediate students he tried to make the subject as simple as possible. He always avoided technicalities, and presented the fundamental facts of Botany in a way calculated to excite the natural curiosity of his students and to awaken and cultivate in them an increasing love for plants. His lectures were brief but inspiring, and the M.Sc. students were never tired of hearing him. It was, however, in the practical class that he made himself felt most. He would

draw the most complicated figures on the blackboard with remarkable ease. His experience as a technician prompted him to make an excellent set of slides of all kinds for class use, and he took delight in seeing and showing what he taught in his lectures. In trips he showed himself to be more active and hardy than his students who were much younger in years. While others would merely look on, he would climb up a most difficult tree with considerable ease, get the things wanted and place them in the hands of his students. I remember a trip in which he took out his M.Sc. students for a study of the aquatic vegetation of a lake. While he was with me and two others standing knee-deep in water and showing at what depths the different plants occurred and how to look for them, some of the students were merely looking on from a distance lest they might spoil their clothes in the water. This brought a well-deserved rebuke which they may remember even now. He never failed to insist that the subject has other value than mere passing of examinations, and always attached greater importance to an actual study of the plants themselves than mere book-reading and the cramming of notes.

While the number of his contributions is not very large, I know that he had a good deal of unfinished work which may now never come to the notice of others. Like several other people, he would start on a problem but after finding out enough about it to satisfy his curiosity, he would give this up and pass on to a different one. Besides, he spent most of his life in an Intermediate College which could not offer much incentive or facility for research, and his duties left him little time for his work. For some time past he was tired of his administrative work and longed to have more time for his research. During his life-time he published a number of works of which a brief account will be given here.

His first Botanical contribution was on "A study of the variation of the number of ray flowers of certain Compositae", published in the Proceedings of the Iowa Academy of Science, Volume 14. This was followed in 1914 by a note on "A method of handling material to be imbedded in paraffin", published in the *Botanical Gazette*, Volume 57. In 1918 he published a detailed account of the Morphology of *Rumex crispus*, published in the *Botanical Gazette*, Volume 66. This work was done at the University of Chicago and formed the subject of his Doctor's thesis. This opened up on him the problem of the Morphological origin of Dicliny and from this time on he began a number of collections

to carry on this work. After his return to India he became more absorbed in studying the ecological aspects of the local vegetation, and this resulted in a valuable paper entitled "A Contribution to the Ecology of the Upper Gangetic Plain", published in the *Journal of Indian Botany* in 1920. Little more than division into phytogeographic regions had been done for India, and his paper is a valuable addition to Indian Botany. In 1922 he was honoured with the Presidentship of the Botany Section of the Indian Science Congress, and he gave a very thoughtful and stimulating address on "The Botanical Opportunity in India". In this he surveyed the Botanical work done in India up to that time and suggested new lines of work. Younger Botanists might even now read this address with profit, since a great deal of what he said at that time remains true even to this day. He used to spend his summer and Dasehra holidays at Mussourie, and in 1923 he published a paper entitled "Succession of Epiphytes in the *Quercus incana* forests at Landour, Western Himalayas", in the *Journal of the Indian Botanical Society*. During this time he and Dr. L. A. Kenoyer (now in the Western State Normal School, Kalamazoo, Michigan, U.S.A.) used to go out on long trips into the Himalayas and made extensive collections and notes. Their results were published in a joint paper, "The Ecology of Tehri Garhwal", in this Journal in 1925.

In 1922 he published his "Guide to Intermediate Botany", an outcome of 10 years' experience with Indian students and 8 years of development and rearrangement of an original six-page laboratory outline. This has been used with considerable success by several teachers of Botany in these Provinces. For some time past he had intended to write an enlarged edition of this work, but for the last 4 or 5 years he was struggling against time and could not devote as much time to his botanical work as he wished to.

As he had spent several summers at Mussourie, he was very well acquainted with the flora of the place and his book "Keys to Mussourie Plants", published in 1929, can be used with advantage by anyone who wants to identify the plants quickly with the unaided eye, even if he has no previous knowledge of Botany. About a year before his death he told me that in the next edition he would put in some more details so that it could be more useful even to serious students of Botany. He had a similar key in preparation (based on leaf characters alone) for the identification of the trees and shrubs of the Upper Gangetic Plain, and parts of it were already typed out when I saw it last.

During the last 4 years he had become greatly interested in the morphology of the mango and wanted to know the reasons why so many flowers fell off without producing any fruit. In a letter, dated February 5, 1928, he wrote—"The mango has become a fascinating problem. It is the most refractory material I have ever tried to work with. But it has the finest kind of 'Degenerationserscheinungen' imaginable. To the best of my knowledge the morphology of mango, and of most of the Anacardiaceae, is unknown".

On the 10th of March 1932, he wrote : "I seem to be excessively busy these days. We are in the midst of our amalgamation proceedings, and association with the University for degree in Agriculture. I get involved in all of it, and it all takes time. I can hardly find time to get some of my Anacardiaceae material rounded up. Have a pretty good supply of four spp. now—*Mangifera, Odina, Buchanania* and *Spondias*—all of great value in connection with the problem of degenerations. Recent cuts of mango seem to suggest that the fusion nucleus is frequently fertilised, while the egg fails to be fertilised. I have made rather large collections to try to follow this out, and will work on it next winter".

On 2nd May, 1932, only a few days before his departure to America on furlough, he wrote : "I am nearing the end of my packing, and can see now how I could finish in a day if necessary. I have a large batch of material of Anacardiaceae in paraffin. Also a host of cakes of *Mangifera*. It will be a joy really to have time and incentive to work and read".

After reaching America he first spent some time in California, then got his family comfortably settled in AMES, Iowa, and himself went over to the University of Chicago. Instead of taking a well-deserved rest as the conditions of his health demanded, he started his work in full swing in the Hull Botanical Laboratory. On 2nd October, 1932, he wrote : "I have a nice room, shared with a man who will take his Doctor's degree in December. I will be here only till the end of December, then spend the rest of the year at Ames. I am specially anxious to be here for the fall quarter, because of the men who will be here then, and not later. Chamberlain is retired, and will spend the time from January to June in Japan, giving a course of Lectures in Tokyo, then after that travelling over the Cycad world as a guest of the Japanese Government. Yamanouchi will be here only the fall quarter also. I can get along without any of the others".

On November 20th, he wrote to another friend "I shall be glad to get home next month and end this perpetual separation from the

family. It is not the way I think I ought to live. Here I am (University of Chicago) still plugging at the mango problem, working hard and making a little progress. But in time it may open up so that finally I may have a contribution that will have a great effect on the botanical world. I feel that I have material here, which if rightly interpreted, will carry a step forward our conception of the development of plants''.

On the same date another friend wrote about him from Chicago. ''Winfield is with us in our cosy little apartment, but we don't see much of him. He is hard at work on his research problem most of the time. He leaves early in the morning and seldom gets home before we are in bed at night''.

These letters show that he was working hard almost up to the time of his death. I understand that upon the death of Dr. C. A. R. Janvier in 1928, he was offered the Principalship of the Ewing Christian College, but like the true devotee of science that he was, he wrote a long letter to the Directors politely declining the offer on the ground that he was better fitted to be a teacher and would have greater satisfaction in being allowed to continue his work as a Botanist. He was, however, persuaded to accept the Acting Principalship till the appointment of a new man, and he heaved a sigh of relief when Dr. C. H. Rice took over the duties in 1930.

The move for an Indian Botanical Society was first initiated by him and the Society which now has a membership of 150 Botanists was brought into being largely by his own efforts and enthusiasm. He was elected its first President and since then he always maintained a keen interest in it. When this Journal was started, it was a very modest and unpretentious undertaking and it often suffered from lack of good articles. Most of the papers were taxonomic notes or floristic descriptions written by a few Botanists who could be counted on one's fingers' ends. In the year 1923, thanks to the efforts of Dr. Dudgeon and some other Botanists, it was taken over by the Indian Botanical Society. Ever since the Journal began to be issued, Dr. Dudgeon determined not to send his papers to any other place for publication, though he all along continued to be a member of the American Botanical Society as well, and might easily have sent his papers to the American Journal of Botany or some other foreign Journal. To his students who were sometimes anxious to have their papers published abroad, he always said ''You are an Indian and should do your best to encourage your own journal''. Today the Journal of the Indian Botanical Society has grown considerably in size and the number

and quality of its papers is a measure of the progress of the study of Botany in this country. It is well known that Dr. Dudgeon contributed a fair share to its development.

Perhaps a scientific Journal like this is not the place to speak too familiarly of him, yet a few incidents of personal contact may not be out of place as they show the real man in him.

I owe so much to him that after I had finished my educational career at Allahabad, I once asked him if I could do anything for him by way of return. Quick came the answer: "Do for your students what I have done for you". I shall never forget that sentence. To me it is pregnant with meaning. I have kept that as my ideal, and have always tried to work up to it. He knew this, and his letters to me showed that he appreciated it.

On one occasion he spoke to me somewhat to this effect; "Panchanan, a Hindu father thinks he has done enough and his life's aim is served if he has a son born and he has been able to give him a good education and has seen him well settled in life. My son is dead (he was killed in a Railway accident in 1923); I wish to leave behind me at least one student who will carry on my name. It will be a great satisfaction if I could push you ahead of me in my life-time". Who knew that he would pass away at a time when I needed him most?

When I went to him for help, he would sometimes keep on with me for long hours and get delayed in his meals. This irritated Mrs. Dudgeon and she never failed to protest, but in this matter she could not bring him round to her way of thinking. During the years 1928 and 1929 he was excessively busy and would seldom find time to look over my work. As he was always disturbed by students and teachers at his bungalow, he would sometimes come over to my room in the hostel after his evening meal and stay till 12 o'clock in the night looking at my slides, offering suggestions and helping in the interpretation of difficult points.

Whenever he was asked for anything he seldom refused. He gave a lot of financial help to several poor students. Some were too poor to purchase books, but used his and retained them for much longer periods than necessary—sometimes several months. This irritated him when it interfered with his own work, but he seldom spoke an unkind word. He was always willing to sacrifice his own time and work for the sake of a sincere student. He was, however, very hard on sluggards and gave expression to his disapprobation in language which though not intended to be offensive, did inflict wounds at times owing to the precision and em-

phasis with which he gave vent to it. Being a strenuous worker himself, he could hardly be expected to tolerate any symptoms of laziness in his students or assistants.

During his life-time Dr. Dudgeon exercised a considerable influence on the Botany of North India and his students are now scattered all over the United Provinces in various Departments of Botany and Agriculture. If he could have had a very few years more of undisturbed work, he could have securely established his reputation as a Botanist in other countries as well. By his death the Indian Botanical Society has lost one of its most enthusiastic members, a man who was loved and admired not only by his students and fellow-workers but by many others in different walks of life. It was his heart's desire to die in India, serving it through the Ewing Christian College. He has left behind him a devoted wife and two daughters. Their grief is the greatest, but they may derive some consolation from the fact that his death is mourned by many others.

PANCHANAN MAHESHWARI.

AGRA COLLEGE,
AGRA,
January 21, 1933.

OBSERVATIONS IN SHORT-CUT TO NECTARY IN
CESTRUM FASCICULATUM, MIRS.

BY

B. N. SINHA.

While spending a holiday in the Nainital Hills, one fair morning my attention was drawn to the visits of a large number of bees hovering round the crimson-coloured bunches of flowers of *Cestrum fasciculatum* growing in my garden. It is a well established observation that bees play a very important role in the pollination of flowers, so *prima facie*, no suspicion was held out with regard to their mischievous activities. But my curiosity was aroused when I saw them sitting at wrong spots namely at the base of the corolla-tube, instead of its open mouth. This fact along with the smallness of the flowers and the bigness of the bees was quite significant of their thievish behaviour. The whole thing thus appeared to be very interesting; so a careful study was made for a period of nearly two months.

During the last two decades or so, the observations in short-cut to nectary has been something of a neglected study. The literature that has accumulated in this direction is mainly due to the labours of Darwin (1), Hermann Müller (7), Fritz Müller Knuth (6) and a few others, based on their extensive work on pollination of flowers in both the hemispheres. Recently Otto Porsch (9) in his admirable work "Vogelblumenstundien" has collected up the literature with regard to the pollination and short-cut to nectaries by birds. Furtado (3) has recorded from Malay, Singapore, Malacca and other places instances of short-cut to nectary by carpenter-bees (*Xylocopa latipes* and *X. aestuans*). In India the observations of such a nature, have been described only in *Sesbania grandiflora* by Tiwary (12), and *Quisqualis indica* L. and *Delphinium sp.* by Iyengar (4). In *Sesbania grandiflora* and *Quisqualis indica*, the cuts are effected by birds while in *Delphinium* by the carpenter-bees (*Xylocopa sp.*). Therefore, from a short review of the available literature, it is apparent that nothing of the kind, has so far been reported with regard to *Cestrum fasciculatum* at least from any part of India.

The Solanaceous genus *Cestrum* is a large one comprising about 150 Species (13) belonging to the Tropical and Sub-Tropical America. The species growing in India, are all exotic ornamental plants cultivated in gardens. But, it is rather remarkable that

some of them have become so well adapted to the climate that they grow very luxuriantly in wild state e.g. *C. elegans, Schlech.* (2) (at Kodaikanal in South India at an elevation of 7,000 ft. above the sea level). *C. Parqui*, L'Herit. (8) (at Abbottabad in the Punjab) and a few others.

Cestrum fasciculatum is a choice beautiful plant of shrubby habit, quite common in Nainital gardens. Usually it flowers from May to December. The flowers in their bud condition, are erect but at maturity they attain something of a nodding position. This appears to be an adaptation to protection of the pollen grains from rain or dew. This fact is already known with certain differences of details in so many plants including even some species of *Cestrum* (5).

The flowers although of very beautiful colour are scentless. The calyx is tubular provided with five teeth and is 8 mm. (tube 5 mm. and teeth 3 mm.) long and 3 mm. in diameter. The corolla is also tubular but it widens out slightly from the base upwards and gets narrowed down again like a barrel before ending into five teeth (fig. 5). Seldom only four teeth are present. The diameter of the corolla-tube at the base is 2 mm. and in its broadest region is 6 mm. The circular opening of the corolla-tube is 3–4 mm. wide and the average length of the tube is 25 mm. The nectary disc which secretes honey, is situated just below the ovary (fig. 4). A fully mature nectary disc secretes so much of honey as to fill about 1/5 of the corolla-tube with the sweet fluid. Certainly is the description indicative of the fact that the plants in their natural habitat are visited by intelligent pollinating agents with long proboscis or small birds (like sun-birds or humming birds) provided with thin long beaks.

The bee which was observed cutting the corolla-tube is *Bombus haemorrhoidalis*, Smith. (Hymenoptera : Apidae). It measures 17 mm. long and 8 mm. broad (Photograph). It cuts holes on the corolla-tube by the help of its two strong jaws and drinks away the honey by inserting its tongue (about 7 mm. long) through the cut or cuts. In habit it is rather timid, as the very presence of man scares it to the extent that it takes to its wings. Due to this, it was found almost impossible to watch from a short distance, the process of cutting the hole in the corolla-tube. It was, however, thus made possible only from a fair distance by the help of a binocular. With great difficulty a few of them could be caught for identification.

These bees usually visit the flowers in the morning, only when it is bright and fair. They come flying about and after making a few rounds near the shrub they sit on one of the branches and select out only those flowers for their attack which are fully grown or nearly so. The young flowers or the buds are not bored at all because they do not find any honey there.

Judging from the description of the flowers and its size (mouth diameter 3–4 mm., length 27 mm.) and that of the *Bombus haemorrhoidalis* (broad 8 mm., long 17 mm.) it appears that these bees could never be expected to be of any use in pollinating the flowers. Being bigger in size they could not even enter the corolla-tube. However, in case of the cuts on the corolla-tube shown in figure 1, it may be possible for the bee to effect self-pollination or even cross-pollination if like-cuts existed on other flowers also, as in this case both the anther and the stigma were accessible to the insect; but in all probability it was only by chance that such cuts were made and that, these had nothing to do with the skill of the bees. The pollinating agents of these flowers must be some other insect or insects with long proboscis or small birds. Attempts to find out the real pollinators have, so far, been futile. Probably very few of these pollinators or perhaps none at all, visit the flowers, because of their being deprived of the honey by *Bombus haemorrhoidalis*. Thus the flowers suffer, and do not get pollinated, with the result that most of them dry up without producing any fruit.

Amongst the flowers examined most of the cuts were situated just above the calyx-tube i.e. at the nearest accessible region of the honey. The following data will give a clear idea about the situation of the cuts:—

Total number of the flowers (with cuts) examined ... ... 200.

1. Flowers with cuts on the calyx-tube as well as on the corolla-tube near the cleft of the calyx teeth (fig. 3) ... ... 8 i. e. 4%
2. Flowers with one cut on the corolla-tube near the cleft of the calyx teeth (fig. 5) ... ... 80 i. e. 40%
3. Flowers with two cuts on the corolla-tube near the cleft of the calyx teeth ... ... 4 i. e. 2%
4. Flowers with long slit-like tortuous cuts beginning from the middle of the corolla-tube and ending in the neighbourhood of the cleft of the calyx teeth (fig. 2) ... ... 16 i. e. 8%

5. Flowers with one cut only in the middle of the corolla-tube (fig. 6)   ...   .   4 i. e.   2%

6. Flowers with one cut in the middle of the corolla-tube and another near the cleft of the calyx teeth (fig. 7)   ...   .   12 i. e.   6%

7. Flowers with one cut in the upper part of the corolla-tube and another near the cleft of the calyx teeth   ...   .   4 i. e.   2%

8. Flowers with three cuts on the upper part of corolla-tube and a fourth near the cleft of the calyx teeth (fig. 1)   ...   .   4 i. e.   2%

As already pointed out, about 1/5 of the corolla-tube is full of honey; and thus the insects which seem to be clever usually make punctures on the tube in a region near the cleft of the calyx-teeth. This process ensures their getting the honey with absolute certainty and also avoids the extra trouble of cutting the calyx as well as the corolla-tube. It will be seen from the data given above that only 4% of the flowers had cuts of the latter nature. For these, the inexperience of the insects is solely responsible. The most common cut was the one situated just in the neighbourhood of the cleft of calyx teeth. Amongst the flowers examined, almost all of them had such cuts with a few exceptions. It is from these that they get an easy supply of the honey. For the cuts in aberrant positions on the tube there can only be two possibilities: these may be due either to the inexperience of some of the foolish *Bombus* or may just be trial experimental searches after the honey before they could locate the right seat of nectar. Any way the fact remains that most of the cuts were very properly situated and that only such flowers which had enough of honey (in almost complete exclusion to the young ones) were the target of attack. This reflects very markedly on the cleverness and intelligence of the insect-thieves. The cuts, however, themselves are not of uniform shape and size (fig. 8). In form they have the outline of amoeba in different stages of its progress as seen under the microscope.

The natural effect of this stealing process of honey certainly has resulted in keeping away the real pollinators or in at least minimising the chances of attracting the right sort of pollinators to visit the newly acclimatised American *Cestrum*. As a consequence of this robbery, the plant is unable to set any seeds and if it is not aided by human agency to propagate on its species by cuttings, such a plant is indeed exposed to the perpetual danger of extinction in its adopted habitat.

Acknowledgement.

I am much thankful to Prof. P. Parija, M.A., I.E.S., for his kindly reading through the MS.; to Mr. T. C. N. Singh for his suggestions during the course of the preparation of this paper; and to Dr. B. Prasad of the Zoological Survey of India for kindly identifying the bee.

BOTANY DEPARTMENT,
RAVENSHAW COLLEGE,
CUTTACK.

Bibliography.

1. DARWIN, C :—*Correspondence in Nature on the subject between* 1874 *and* 1877.

2. FIRMINGER'S *Manual of Gardening for India* (6th. Edition), 1918.

3. FURTADO, C. X.:—Note Short-cuts to Nectaries by Xylocopas, *Journal Indian Botanical Society*, Vol. IV, No. 1, 1924.

4. IYENGER, M. O. P.:—Two instances of Short-cuts by animals to the Nectaries of Flowers. *Journal Indian Botanical Society*, Vol. III, Nos. 9 and 10, 1923.

5. KERNER, A. VON MARILAUN:—*The Natural History of Plants* (1904) Part II (Eng. Translation by F. W. Oliver).

6. KNUTH, P :— *Handbook of Flower Pollination* (Eng. Trans. 3 Vols.)

7. MULLER HERMANN :—*The Fertilization of Flowers* (Eng. Trans.)

8. PARKER, R. N.:—*A Forest Flora for the Punjab* (1918).

9. PORSCH OTTO:—Vegelblumenstudien II, *Jahrbücherfür wissens cheftliche Botanik*, Seibenzigster Band, Zweite Heft 1929.

10. SWYNNERTON, C. F. M..—On Short-cuts by birds to Nectaries. *Journal Linn. Soc. Bot.* Vol. XLII, 1916.

11.   „   Short-cuts to Nectaries by Blue Tits. *idid.*

12. TIWARY, N. K.:—A note on a Short-cut to the honey in *Sesbania grandiflora. Journal Ind. Bot. Soc.*, Vol. V, No. 3, 1926.

13. WILLIS, J. C.:—*A Dictionary of the Flowering Plants and Ferns* (Cambridge University Press), 1919.

Explanation of the Plate.

(c=calyx-tube, p=corolla-tube, a=anther, s=stigma, n.d.= nectary-disc, c. c.=cut on the calyx-tube, c. p.=cut on the corolla-tube)—The 'cuts' have all been drawn by the help of a Zeiss Camera lucida.

FIGURES 1-3 and 6-7: Portions of flowers showing the different positions of the cuts in relation to the calyx- and corolla-tube. × 4.

FIGURE 4: A longitudinal section of the flower. × 2.

FIGURE 5: A flower of *Cestrum fasciculatum*. Showing the commonest type of cut. × 2.

FIGURE 8: Ten different types of cuts on the corolla-tube. × 4.
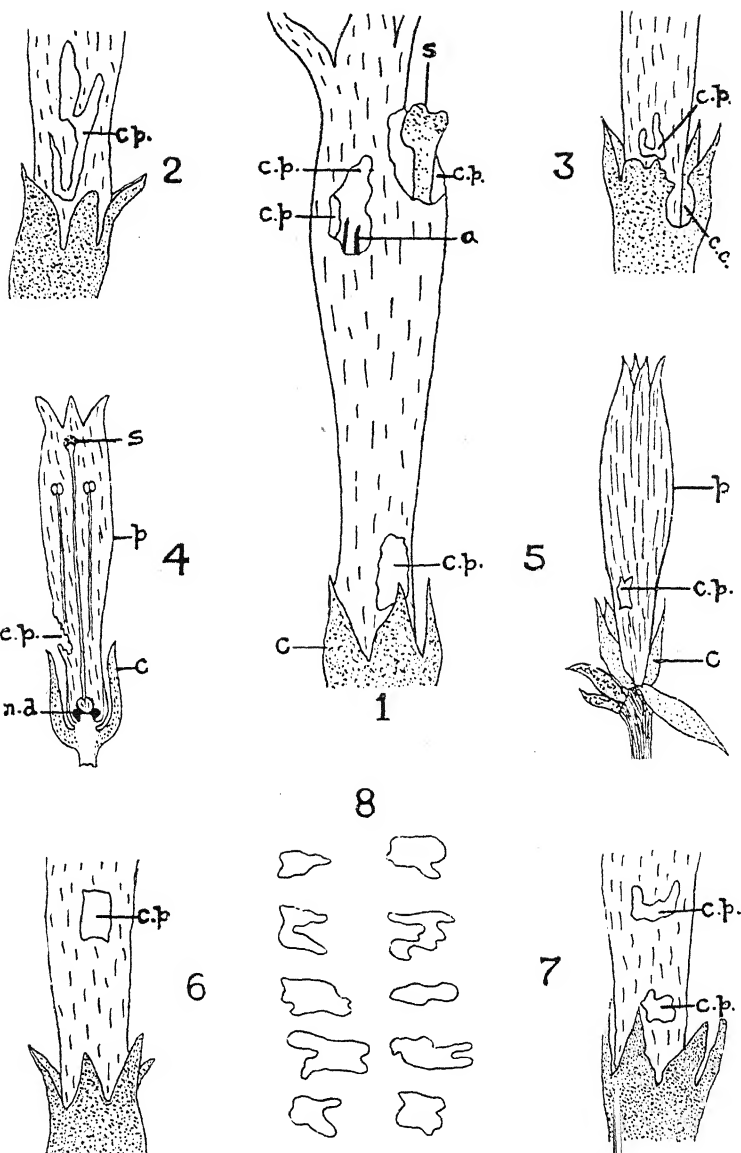
Bombus haemorrhoidalis. Smith. Showing the insect from three different views a = dorsal view, b = side view, and c = ventral view.　1½.

MEGASPORE FORMATION AND EMBRYO-SAC OF *ARGEMONE MEXICANA*, LINN.

BY

A. C. JOSHI,

Department of Botany, Benares Hindu University, Benares.

With one plate and one figure in the text.

Introduction.

Argemone mexicana Linn. is one of the first American plants to be naturalised in India. The exact date of its entrance into our country is unknown, but that it was certainly here, even in the beginning of the 19th century, is proved by a reference to the works of Roxburgh (14) and Wight and Arnott (25). At the present time it is one of the commonest weeds all over the country.

Last year, the writer collected some plants of *Argemone mexicana* from a place in Punjab near Hoshiarpur bearing flowers quite different from the normal. Their external morphology was described sometimes ago (10). Later on, it was decided to study their anatomy too, the results of which it is hoped will be published shortly. For a proper appreciation of the structure of these abnormal flowers, however, it was thought necessary to work out the structure of the normal flowers also. Consequently, parts of two normal flowers were fixed and embedded in February last and later on microtomed. Some of these slides also yielded several good sections of the ovules, showing the formation of megaspores and the embryo-sac. On consulting Schnarf (15) and Schürhoff (16), it was found that nobody has so far studied the megasporogenesis or any other phase of the life history of this species and as it was found to show some important differences from the structure normal for the flowering plants, it was considered worth while to publish the results.

The Material.

The material of *Argemone mexicana* used in the present investigation was collected at Benares from the Hindu University grounds. It consisted of two ovaries, one from an unopened bud and the other from a flower which was in full bloom. It was trimmed from the sides to remove the various prickles and the two ends to facilitate the penetration of the fixative, which was a mixture of formalin, glacial

acetic acid and 60 per cent alcohol in the proportion of 5 : 5 : 100. Sections were cut from one half of each ovary in a transverse manner and from the other half longitudinally. A combination of Safranin and Gentian Violet was used in staining.

Megaspore Formation.

The development of the megaspores was studied from sections provided by the younger ovary. This is illustrated in figures 2, 3, 4 and 5. In this ovary the megaspores were already formed, so that the earlier stages were not seen, but so far as figure 3 can give any indication, it appears that there is only a single hypodermal archesporial cell which divides by a transverse wall to form the primary wall cell and the megaspore mother cell. The wall cell appears to divide once anticlinally and once or twice periclinally to form a portion of the wall, 2 to 3 cells thick, which separates the sporogenous tissue from the epidermis of the nucellus. The megaspore mother cell, as usual, divides first by a periclinal wall into two cells, one upper and one lower. In the majority of flowering plants, both monocotyledons and dicotyledons, both these cells divide again in the same manner by periclinal walls to form a linear row of 4 megaspores. In the case of *Argemone*, however, their behavior is different. Here the upper cell divides first by an anticlinal wall to form two megaspores arranged in a line transverse to the long axis of the ovule. The division in the lower cell begins later than in the upper cell. This can be clearly seen from figures 2 and 4. Further the division wall here is periclinal, i.e., in a plane at right angles to the division wall of the upper cell and gives rise to two megaspores arranged longitudinally. The tetrad thus formed is not of a linear type, but "T-shaped", to use the terminology of Palm (11), who was the first person to give a complete classification of the different types of megaspore formation in the flowering plants.

Sometimes in longitudinal sections of the ovule, the megaspore mother cell appears to give rise only to 3 megaspores (figures 4 and 5). This is, however, only due to the plane of the section. If longitudinal sections of the ovule are cut in a plane at right angles to that of figures 2 and 3, i.e., in a more or less tangential longitudinal plane, only one of the two upper megaspores is seen at a time and it appears that there are only 3 megaspores.

The morphology of the family Papaveraceæ, to which *Argemone mexicana* belongs, has, on account of the relations of this family with the cruciferæ and the peculiar structure of the flower of two sub-families Hypecoideæ and Fumarioideæ, been fairly extensively studied. Schnarf (15) cites more than 20 contributions dealing with the

development and life history alone of the various members; and although nothing has been so far published about the morphology of the genus *Argemone*, we have a fair knowledge about the genera *Eschholtzia, Hypecoum, Chelidonium, Sanguinaria, Glaucium, Papaver, Dicentra, Corydalis* and *Fumaria* due to the works of Shaw (17), Huss (9), Hegelmaier (5), Surface (20), Vesque (22), Hofmiester (7), Warming (24), Guignard (4), Tischler (21), etc. A summary of all the work is given both by Schürhoff (16) and Schnarf (15), but a 'T-shaped' tetrad of megaspores has not been described in any of these genera. In this respect *Argemone* appears to be different from all of them and resembles such plants as *Butomus* described by Ward (23), *Jeffersonia* described by Andrews (1), *Potamogeton* by Holferty (8), *Diospyros* by Yasui (26), *Trillium* investigated by Heatley (5) and Spangler (18) and *Thismia* and *Gasteria* studied by Pfeiffer (12, 13) and Stiffler (19) respectively. There appears to be, however, no particular phylogenetic significance in this type of megaspore tetrad as it is found in families so wide apart as the Butomaceæ, Berberidaceæ, Potamogetonaceæ, Ebenaceæ, Liliaceæ, Burmanniaceæ and Papaveraceæ and its presence in any one plant is no guide to its affinities. Its importance lies in its serving as a link between the tetrahedral tetrad in which the pollen grains are usually arranged and the linear tetrad which is the usual form of megaspore tetrad and bridging the gap between these two extreme types.*

Of the 4 megaspores formed in *Argemone mexicana*, as is normally the case in flowering plants, the three upper ones soon degenerate after their formation and the lower one alone functions and develops into the embryo-sac (figures 2, 3 and 4). In one case, however, a deviation was noticed from this rule. Here (figure 5) it was the second megaspore from below which had remained functional, while the lowermost along with the two upper ones had degenerated. Such variations are known to occur in other plants also and a description of these, as were known in their time, is given by Coulter and Chamberlain (3) on pages 84 to 86; and as they put it, these serve to emphasize the megaspore character of all the four cells of the tetrad.

---

* The following other genera of plants are mentioned by Schnarf (Embryology der Angiospermen, Berlin, 1927; pp. 97-98), in which a 'T-shaped' arrangement of the megaspores has been recorded :

Urtica, Cynomorium, Allionia, Brasenia, Cabomba, Drimys, Asimina, Adonis, Ranunculus Myosurus, Sarracenia, Garcinia, Moringa, Saxifraga, Neurada Hydrostachys, several Cistaceæ, Malvaceæ, and Tiliaceæ, *Daphne, Cortusa, Cynoglossum, Erythraea, Gentiana, Villarsia; Asclepias, Valeriana, Triglochin, Ruppia, Tricyrtis, Yucca*, several Amaryllidaceæ, *Burmannia, Heteranthera, Typha* and *Platanthera*.

Embryo-Sac.

The structure of the embryo-sac was studied from sections provided by the older ovary. Owing to its large size it could not be possible to get perfect sections, showing all the parts in the same. For this reason a reconstruction was made from a number of serial sections and it is reproduced in the accompanying text-figure (fig. 1). This figure represents a longitudinal section of the ovule and show
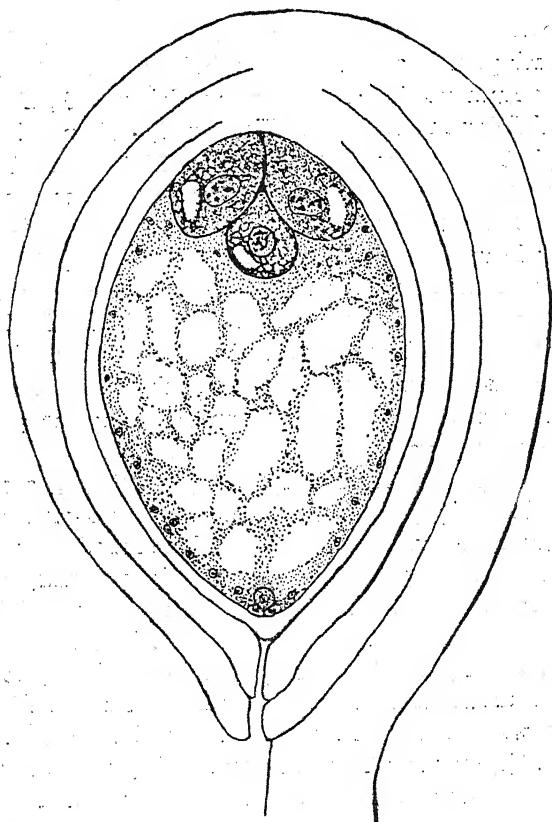
FIG. 1. *Argemone mexicana*. Longitudinal section of the ovule—a reconstruction from a number of serial sections, showing the great disparity in the size of the egg-apparatus and the antipodals. The endosperm formation has set in and the synergids have degenerated. × 100.

the structure of the ovule when endosperm formation had already set in. Sections showing a stage earlier than this were not available, except a 2-nucleate embryo-sac which was provided by the younger

ovary and is sketched in figure 3, but it seems to be fairly certain from the study of the egg-apparatus and the antipodals that the normal '8-nucleate type' of embryo-sac is formed.

The chief feature of the embryo-sac is the great difference in the size of the cells of the egg-apparatus, namely the egg cell and synergids and the antipodals. The latter are about 8 to 10 times larger than the former. The egg-cell is just about 25 *microns* in diameter. The synergids are even smaller than this. They soon get disorganised and do not appear to take any part in the process of fertilisation. The antipodals, on the other hand, at this stage are about 200 *microns* in diameter and form the most conspicuous part of the embryo-sac. Their detailed structure is shown in fig. 6. At this time, disorganisation has already set in, as can be clearly seen from the appearance of the nuclear wall. The protoplasm is of an alveolar nature and in the nucleus, its wall is breaking down. The nucleolus has broken up into a number of darkly staining masses which seem to be in a process of gradual dissolution. In some of these nucleolar masses small shining bodies could be seen. Their exact nature has not been studied by the writer but these appear to be of the nature of dissolution products.

Another feature of the antipodals is the presence of large vacuoles inside them. These are clearly shown in Fig. 6 which represents a transverse section of the antipodals. The vacuoles seem to occupy a more or less definite position. In a transverse section these are found towards the outside of the nucleus of every antipodal cell and on the side away from the centre of the embryo-sac. In longitudinal sections of the ovule, the antipodal cells show a vacuole toward their anterior end facing the micropyle. It thus appears that every antipodal cell has got a vacuole which starts from the micropylar anterior end of the cell and extends peripherally on to the side away from centre of the embryosac.

In the above structure of the antipodals, *Argemone mexicana* shows perfect resemblance with the other Papaveraceæ whose structure is known. Schürhoff (16) says that the "Papaveraceæ are characterised by large antipodals. These are uninucleate, in groups of three and possess a vacuole in the anterior plasma." The embryosac of *Argemone* agrees in every way with the above characterisation. Here, however, we know further that the vacuoles, which are certainly present in the anterior part of the plasma of the antipodals, extend into their sides also.

Huss (9), who studied the antipodals of *Fumaria, Corydalis* and *Papaver*, found that the antipodals are not very big in the young condition but these enlarge to a very great extent by the time these

begin to degenerate. The antipodals of *Argemone* studied by the writer were in early stages of degeneration. He did not see any young antipodals and it is not possible to say, whether the antipodals in this plant are very large in size from the very beginning or these attain this large size only at the time of their degeneration. The degeneration stages of the antipodals of *Argemone* show great resemblance with similar stages figured by Huss in his material.

Another feature appearing to be peculiar to the embryosac of *Argemone mexicana* is the very early and quick development of the endosperm. The formation of endosperm had set in all ovules of the older ovary examined by the writer whether there was a trace of the penetration of the pollen tubes or not and fertilisation had taken place or not. In figure 1, for instance, the egg has not yet divided when a good deal of endosperm has been formed. This leads one to suspect that here also the formation of the endosperm may be beginning even before fertilisation, as reported by Coulter (2) for *Ranunculus*. The very near position of the two families in every recent system of classification of flowering plants lends a still further colour to this suspicion But, whether this is true or not, this much is certain, that a great deal of endosperm is formed before the first division of the oospore and the beginning of embryo-formation.

The pollen-tube was seen to enter through the micropyle in all the cases observed.

A few early stages in the formation of embryo were seen in this ovary, but these were too few to enable one to give any description.

Summary.

1. The megaspores of *Argemone mexicana* form a ' T-shaped ' tetrad.

2. The embryosac is characterised by very large antipodals which are 8 to 10 times as large as the egg-cell. They have got large vacuoles running from their anterior ends on to the outer sides. The synergids are even smaller than the egg-cell and degenerate very soon. A great deal of endosperm is formed before the first division of the oospore.

BENARES,
20th August 1932.

Literature Cited.

1. ANDREWS, E. M.—Development of the Embryosac of *Jeffersonia diphylla*. Bot. Gaz. 20 : 423-424, 1895.

2. COULTER, J. M.—Contribution to the Life History of Ranunculus. Bot. Gaz. 25 : 73-88, 1898.

2. COULTER, J. M. & CHAMBERLAIN, C. J.—Morphology of Angio-
sperms. New York, 1903.

4. GUIGNARD, L.—La formation de L'embryon chez l'Hypecoum.
Jour. Bot. 18 : 33–44, 1903.

5. HEATLEY, M.—A study of the life history of *Trillium cernuum*.
Bot. Gaz. 61 : 425–429, 1916.

6. HEGELMAIER, F.—Vergleichende Untersuchungen über die Entwik
—lung dikotyledoner Keime mit Berucksi-
chtigung der pseudo-monokotyledonen. Stutt-
gart, 1878.

7 HOFMEISTER, W.—Neuere Beobachtungen über Embryobildung
der Phanerogamen. Jahrb. Wiss. Bot. 1 ;
82–188, 1858.

8. HOLFERTY, G. M.—Ovules and embryo of *Potomogeton natans*.
Bot. Gaz. 31 : 339–346, 1901.

9. HUSS, G. A.—Beiträge zur Morphologie und Physiologie der
Antipoden. Beih. Bot. Centralb. 20 : 77–174.
1906.

10. JOSHI, A. C.—Abnormal Plants of *Argemone mexicana*. Proc.
Ind. Sci. Cong. Bangalore, 1932, (abstract).

11. PALM, B.—Studien über Konstruktions-typen und Entwick-
lungswege des Embryosackes der Angiospermen
Diss. Stockholm, 1915.

12. PFEIFFER, N. E.—Morphology of *Thismia americana*. Bot. Gaz.
57 : 122–135, 1914.

13.——————The sporangia of *Thismia americana*. Bot. Gaz.
66 : 354–363, 1918.

14. ROXBURGH, W.—Flora Indica. Calcutta, 1874.

15. SCHNARF, F.—Vergleichende Embryologie der Angiospermen·
Berlin, 1931.

16. SCHURHOFF, P. N.—Die Zytologie der Blutenpflanzen. Stuttgart,
1926.

17. SHAW, CH. H.—Note on the sexual generation and development of
the seed-coats in certain of the Papaveraceæ.
Bull. Torr. Bot. Club. 31 : 429–433, 1904.

18. SPANGLER, R. C.—Female gametophyte of *Trillium sessile*. Bot.
Gaz. 79 : 217–221, 1925.

19. STIFFLER, E. G.—Development of embryosac in *Gasteria, Cyrtan,
thus* and *Velihemia*. Bot. Gaz. 79 : 207–216,
1925.

20. SURFACE, F. M.—Contribution to the history of *Sanguinaria Canadensis.* Ohio nat. 6 : 379–385, 1905.

21. TISCHLER, G.—Untersuchungen über die Entwicklung des Endosperms und der Samen-schale von *Corydalis cava.* Verh. med.—Nat. Ver. Heidelberg. 6 : 351–380, 1900.

22. VESQUE, J.—Nouvelles recherches sur le dévelopement du sac embryonnaire des phanérogames angiospermes. Ann. Sci. Nat. Bot. VI. 8 : 261–390, 1879.

23. WARD, H. M.—A contribution to our knowledge of the Embryo-sac in Angiosperms. Jour. Linn. Soc. London. Bot. 17 : 519–546, 1880.

24. WARMING, K.—De l'ovule. Ann. Sci. Nat. Bot. VI. 5 : 177–266, 1878.

25. WIGHT, R. & ARNOTT, G. A. W.—Prodromis Floræ Peninsulæ Indiæ Orientalis. London, 1834.

26. YASUI, K.—Studies of Diospyros Kaki. Bot. Gaz. 60 : 362–373, 1915.

Description of the Plate.

All the figures are of *Argemone mexicana* and were drawn with the help of a camera lucida.

Fig. 2. A tetrad of 4 megaspores, the two upper arranged transversely, the two lower longitudinally. The two upper-most have completely degenerated, while the second from below is in the process of degeneration. The lowermost is the functional megaspore. × 1300.

Fig. 3. A part of the longitudinal section of the ovule, showing the upper part of the nucellus and a part of the inner integument. The megaspore tetrad here is older than the one shown in fig. 2. All the three upper spores have completely degenerated while the nucleus of the lowermost functional megaspore has divided to form a 2-nucleate embryosac. × 1300.

Fig. 4. A view of the megaspore tetrad in a tangential longitudinal section of the ovule. Here only one upper spore is seen. The second spore from below has not yet degenerated. × 1300.

PLATE I.   J. I. B. S. XII : 2.
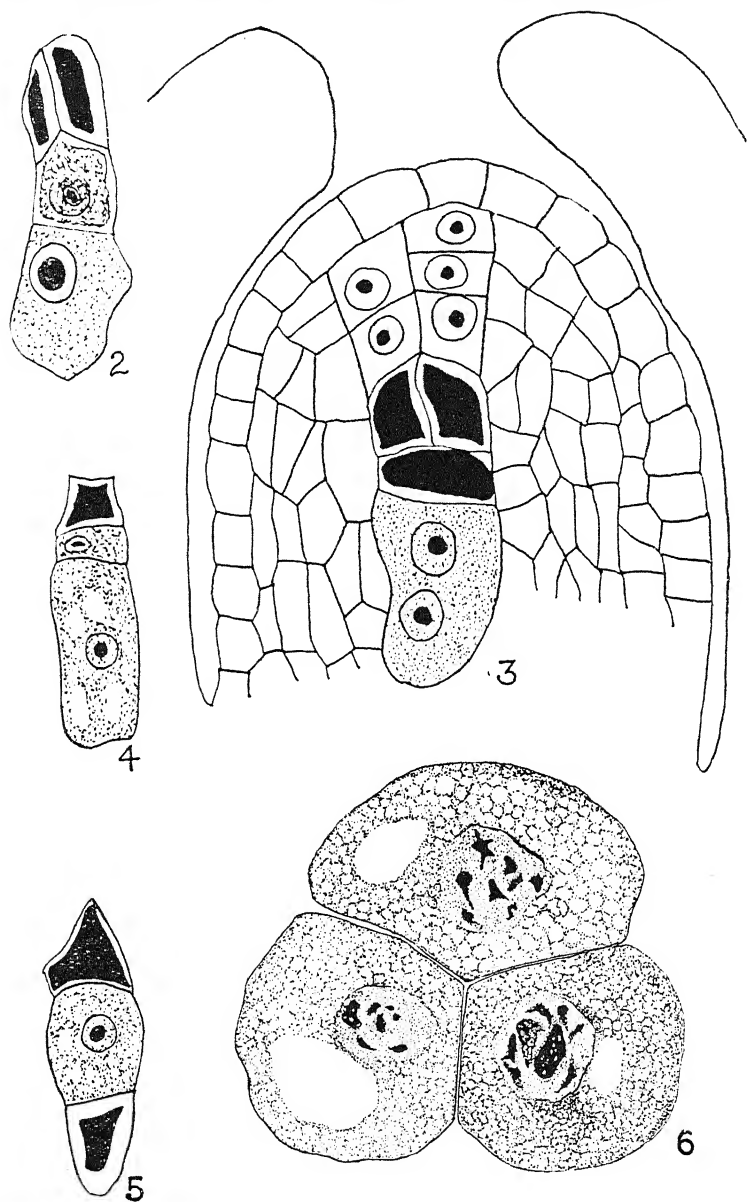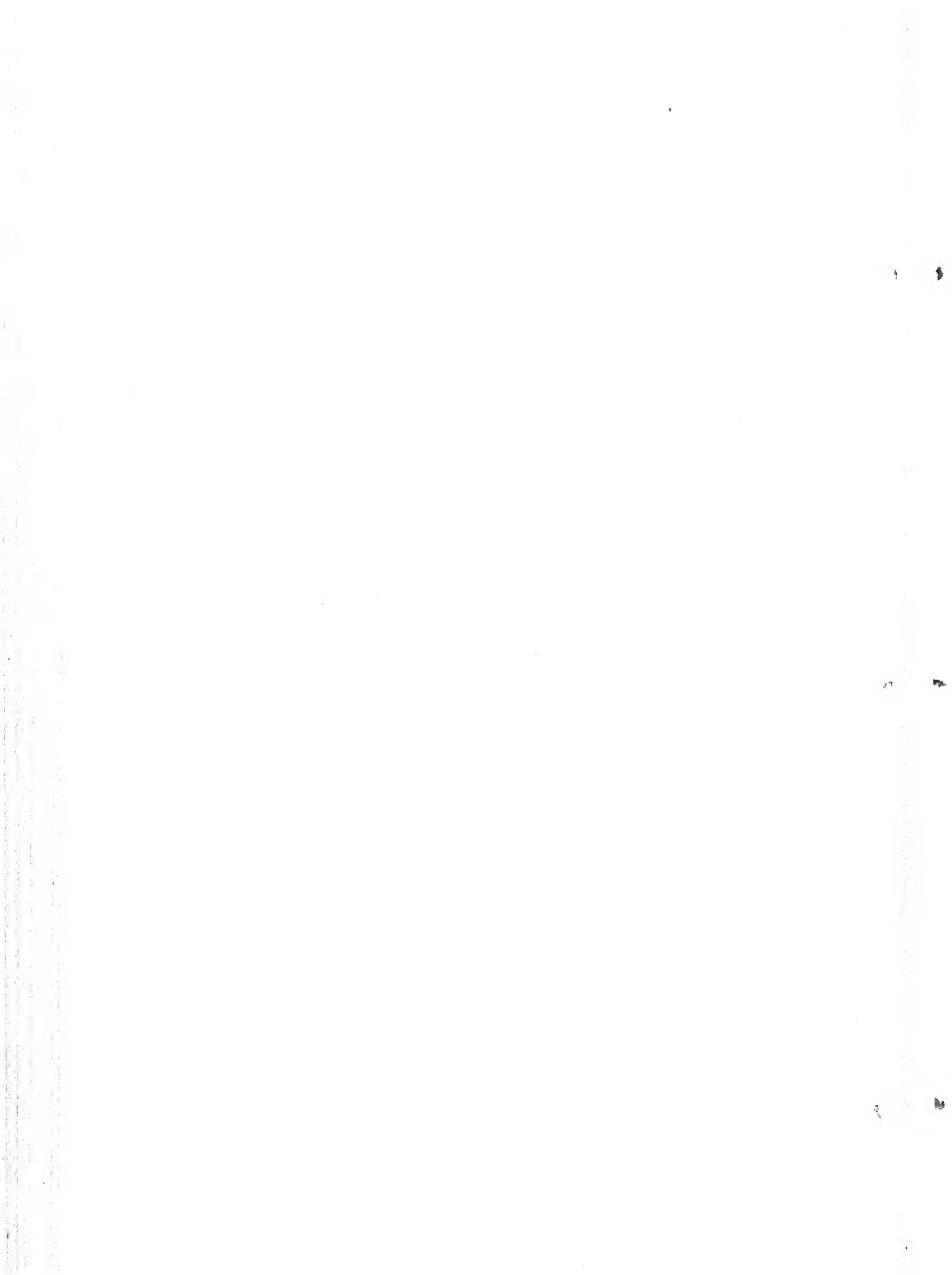
Fig. 5. Same as figure 4. Here, however, the lower-most spore has degenerated and the second spore from below is the functional one. × 1300.

Fig. 6. A transverse section of the three antipodals. These are just beginning to degenerate. The wall of the nucleus is becoming faint, the nucleous has broken up into a number of darkly staining masses which are being gradually dissolved. Each antipodal cell shows a vacuole towards the outside. × 400.

SOME OBSERVATIONS ON THE FLOWERS OF
ERYTHRINA INDICA, LAM.[*]

BY

A. B. SARAN, M.Sc. (AGRA),

Rice Research Department, Sabour.

The observations recorded in this contribution were made as Patna, during the latter part of March, 1932. The author had an occasion to notice four kinds of birds, the common bees, and the big black ants visiting the flowers of one Coral-tree, (*Erythrina indica*, Lam), almost leafless, in its full bloom. The birds were seen flying from one flower cluster to another and appeared to be biting out a portion of the flower and sucking up the honey. This prompted the author to make a close study of these flowers, specially in respect to the visits paid by these birds.

Instances are not uncommon, where flowers have been visited by birds for sucking up the honey. Cases of short-cuts to nectaries are also very common and have been recorded by Furtado [1], Swynnerton [2,3], Tiwari [4,5] Mullers [6], Debbarman [7], Knuth [8], Iyengar [9] and others.

---

[*] The work was done in the Botany Department, Ravenshaw College, Cuttack.

[1] Furtado, C. X. Note Short-cuts to Nectaries by Xylocarpas, Jour. Indian Bot. Soc., Vol. IV, 1924.

[2] Swynnerton, C. F. M. Short-cuts by Birds to Nectaries, Jour. Linn. Soc. Bot. Vol. XLIII, 1916, p. 381.

[3] Swynnerton, C. F. M. Short cuts to Nectaries by Blue Tits, *ibid*, p. 417.

[4] Tiwari, N. K. A note on a Short-cut to the honey in *Sesbania grandiflora*, Jour. Indian Bot. Soc., Vol. V, 1926.

[5] Tiwari, N. K. A note on the Flowers of *Tecoma radicans*, Jour. Indian Bot. Soc., Vol. VIII, 1929.

[6] Muller, H. The Fertilisation of Flowers. Eng. Trans. by Thompson, 1894.

[7] Debbarman, P. M. A short note on the short-cut to the nectar in the flowers of *Castqnospermum australe* C. and F. Proc. Ninth Indian Sci. Congress 1923.

[8] Knuth, P. Handbook of Flower Pollination. Eng. Trans. by Wilson and Davis, 1906-8.

[9] Iyenger, M. O. P. Two instances of short-cuts by animals to the Nectaries of Flowers, Jour. Indian Bot. Soc., Vol. III, 1923.

Swynnerton [10], however, has observed short-cuts to nectaries in two species of Erythrinas, *E. humeana*, Spreng. and *E. tomentosa*, R. Br. In both these cases the damage is done by Sunbirds, Widow-birds and by such other birds, which damage the flowers more or less indiscriminately. In this contribution, the author neither specifies any particular portion or portions of the flower, where the injury was inflicted nor mentions the nature of the damage done.

Ali [11] has enumerated as many as 46 birds that visit the flowers of *Erythrina indica*, Lam for eating nectar. But no mention is made by him, about the damage, if any, done by any of these birds to the flowers. He, however, has observed that the striped squirrels bite into and suck the nectar sac.

To the best knowledge of the present author there is no record of any previous study on short cuts to nectaries in *Erythrina indica*, Lam., so an attempt has been made here to describe it in some detail.

In order to understand the ways by which the birds, which visit the flowers of *Erythrina indica*, Lam., carry out their business a short description* of the interesting features of the flower seems to be necessary.

The coral-red flowers are produced in dense racemes. The wing and the keel petals are almost of equal size. The upper margins of the two wing petals closely overlap each other and their lower margins partially overlap the keel petals, which in turn partially overlap the staminal tube (Fig. 1). Thus a closed pouch is formed (Fig. 2). The honey which is secreted by a number of rod-shaped glands situated at the inner base of the staminal tube (Figs. 3 & 4), is collected at the anterior end of this pouch.

Just after the flowers open (at mid-night) they contain very little honey. The quantity of the honey increases with the time and the following evening the maximum is reached. The honey is sweet with a slightly bitter taste. Two samples of the honey were collected in the evening and on analysis they were found to contain 26-27 per cent of reducing sugar.†

---

[10] Swynnerton, C F. M. Short-cuts by Birds to Nectaries, Jour. Linn. Soc. Bot. Vol. XIII, 1916, p. 381.

[11] Ali, Salim A. Flower-birds and Bird-flowers. Jour. Bom. Nat. His. Soc. Vol. XXXV, No. 3, February 1932.

* For a fuller description the readers are referred to any book of Systematic Botany.

† The honey was collected at Patna in a dry clean glass-stoppered bottle to which a little of toluol was added to prevent bacterial growth. It was analysed after 72 hours from the time of collection.

While the common bees and the big black ants visit the flowers for pollen and honey, the following birds were seen biting out a little hole at the anterior end of the honey pouch and sucking up the honey of the flowers of this plant :—

1. Crateropus canorus	...	The Seven Sisters.
2. Arachnecthra asiatica	...	The Sunbird.
3. Acridotheres tristis	...	The Common Myna.
4. Acridotheres fuscus	...	The Jungli Myna.

The most common visitors among these birds are the Seven Sisters. They visit the flowers from morning to evening. The Sunbird comes at about 8 o'clock in the morning and the two types of Mynas are the occasional visitors. All these birds come in search of honey. Since the honey is stored in a closed pouch, it has to be 'rifled' or a little hole has to be cut out before the honey can be reached. The Seven Sisters and the Mynas bite out a little hole (Fig. 1) at the anterior end of the honey-pouch, while the Sunbird by means of its fine beak 'rifles' a little opening at the anterior end of the honey-pouch, in order to sip off the honey.

It is interesting to find that all these birds either bite out or 'rifle' a little hole at the anterior end of the honey-pouch, where the honey collects, and not anywhere else.

The other visitors are the common bees and big black ants. The bees come in search of pollen grains, which they take away in large quantity. They do not seem to make any attempt to get at the honey. The ants were seen entering and coming out of the honey-pouch through the holes cut open by the birds. Most likely they visit the flowers for the honey and seems to feed upon what is left behind by the birds.

While making these observations the author's attention was drawn to the pollination of these flowers. Singh[12] has observed at Lucknow that Mynas *(Acridotheres tristis)* promote pollination in *Erythrina indica*, Lam. According to the present author's observations at Patna the most common visitors to the flowers of *Erythrina indica*, Lam., are the Seven Sisters. These were seen flying straight from one bunch of flowers to another in large numbers throughout the day in search of honey. Judging from the frequency of visits paid by these birds, it may be said, that in this case, the main credit for promoting pollination goes to them.

---

[12] Singh, T. C. N. A preliminary note on the Pollination of the Coral-tree *Erythrina indica*, Lam.) Jour. Bombay Natural History Soc., Feb. 15, 1929.
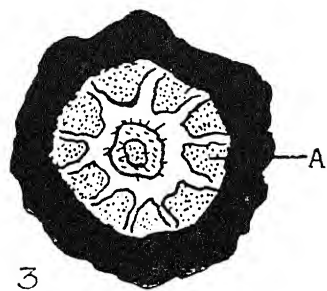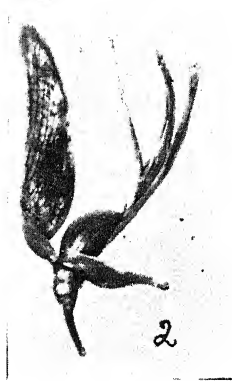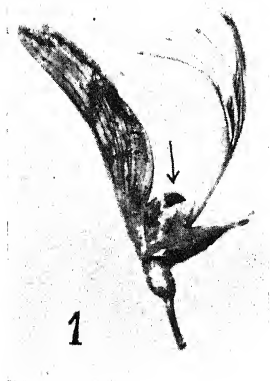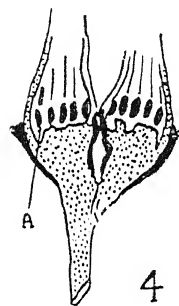
Plate I.

Explanation of Figures.

Fig. 1. Photographs of the flower, showing the hole (at the tip of the arrow), which has been made by the birds at the anterior end of the honey-pouch $\times \frac{3}{4}$

Fig. 2. Photograph of a flower, showing the uninjured pouch $\times \frac{3}{4}$

Fig. 3. Camera lucida drawing of a Transverse Section at the basal region of a flower, showing the honey secreting glands at A. $\times 8$

Fig. 4. The lower portion of a flower cut open longitudinally to show the position of the honey secreting glands, A. $\times 2$.

EXPLOSIVE FRUITS IN *VISCUM JAPONICUM*, THÚNB.

BY

B. SAHNI,

University of Lucknow.

It is generally believed that the seeds of the genus *Viscum*, and probably of all the species of *Loranthus*, are dispersed by birds, who eat the viscid pulp and leave the seeds sticking to the twigs of trees on which they rub their beaks. Another view which was current for some time, but which has been experimentally shown to be mistaken at least in the case of *V. album*[1]; is that the seeds must pass through the alimentary canal of a bird before they can germinate. There is probably no doubt that the wide distribution of the Loranthaceæ is chiefly due to the viscid nature of the berries which are carried by birds even to distant oceanic islands. For this reason some interest attaches to those members of the family in which the seed, although embedded in a viscid pulp, does not seem to depend upon animals for its dispersal.

The object of the present note is to record one such case, namely *Viscum japonicum*, Thunb., which I have had under observation during a recent summer vacation at Lansdowne in Garhwal (Western Himalayas). In a mixed forest in which the dominant species of trees is *Quercus incana* (associated with *Rhododendron arboreum*, *Pinus longifolia*, etc.) there was hardly an oak which did not show one or more bunches of the parasite. The minute obovoid fruit ripens during the rainy season, which begins in July. The berries[2] are scarcely 2 mm long; when quite ripe they are somewhat translucent, the seed being just visible in the broader distal part of the berry. As the fruit is so small and inconspicuous, and as I never noticed any bird visitors, I kept some plants under close observation with a view to ascertain the mode of dispersal. Within the first fortnight of the monsoon season a number of seeds were found sticking to neighbouring twigs and leaves of the oak, and even on the moss-covered trunks far away from the attacked branches. I imagined at first that a possible agent in the dispersal might be the elusive cicadas, which are active at dusk, and abound in the forest. This was soon proved to be not the case. Oak twigs bearing ripe mistletoes were kept indoors overnight,

---

[1] See Engler and Prantl (1893), p. 160.

[2] The berry of the Loranthaceæ is in reality a false fruit as it includes the hollowed thalamus in which the ovary is sunk (cf. Engler *loc. cit.*).

covered with a handkerchief, the cut ends dipping in water. In the morning a number of seeds were found sticking to the cloth or lying in the water; the empty fruits as a rule remained attached to the *Viscum*, but in a few cases these were also lying free. Later in the season, when ripe fruits were common, I repeatedly confirmed the fact that the seeds are ejected with force to a distance of two feet or more ; they are thus liable to get stuck to neighbouring twigs or fall upon branches lower down or get washed down by the rain. If caught in a gust of wind after being ejected they may even be carried to other trees. Their dispersal through any other agent, such as birds, seems to me very doubtful, except as a chance occurrence, e.g., when a seed during its flight through the air may happen to strike a bird. The ejection of the seeds can be conveniently watched if shoots bearing ripe fruits are kept completely immersed in water. It is then seen that the broad upper end of the fruit becomes very turgid and bursts at the apex, the seed travelling several inches under water.

I am not aware that the explosive mechanism in the fruit has ever been described in this or in any other species of the genus.

Among works on the Indian flora I have seen descriptions of *V. japonicum* by the following authors: Bamber, Brandis, Collett, Engler, Fyson, Hooker, Kanjilal and Parker.[3] None of them refers to the explosive nature of the fruit. Other species of the genus have been described from India, Burma, Ceylon and Java by Cooke, Haines, Kurz, Koorders and Trimen [4]. Neither these authors, nor Bentham and Hooker, nor Engler and Prantl say anything about this feature in *Viscum*; while Kurz even says that the fruit is indehiscent in the whole family. This latter statement is no doubt wrong. Prof. S. R. Kashyap drew my attention to Parker's[5] statement that in *Arceuthebium Oxycedri* (a minute parasite common on *Juniperus* in the interior N.-W. Himalayas) the seed is forcibly ejected in a manner somewhat like that described above. The process was first described by Reinaud de Fonvert in 1845 [6]. The chief point of difference is that in *Arceu-*

[3] Bamber (1916), p. 644; Brandis (1911), p 552; Collett (1902), p. 440; Engler in Engler and Prantl (1893), p. 195; Fyson (1915), p. 356; Hooker (1890), p. 226; Kanjilal (1911), p. 339; Parker (1918), p. 440.

[4] Cooke (1906); Haines (1921), pp. 803-804; Kurz (1377), p. 323; Koorders (1912); Trimen (1895) p. 472.

[5] Parker (1918), p. 440. This author adds, p. 411, that presumably the seeds of *A. minutissimum* are dispersed in the same manner.

[6] Reinaud de Fonvert (1846); see also Stewart (1874), p. 395; Bentham and Hooker (1880), p. 213; Johnson (1888), p. 150, pl. X fig. 8; Engler and Prantl (1893) p. 193; Peirce (1905), p. 90; Brandis (1911) p. 553.

thobium the fruit wall is regularly thrown off as a cap which separates along a basal ring-shaped line of dehiscence. This latter fact is interesting because in *Viscum japonicum*, too, as already stated, the fruit
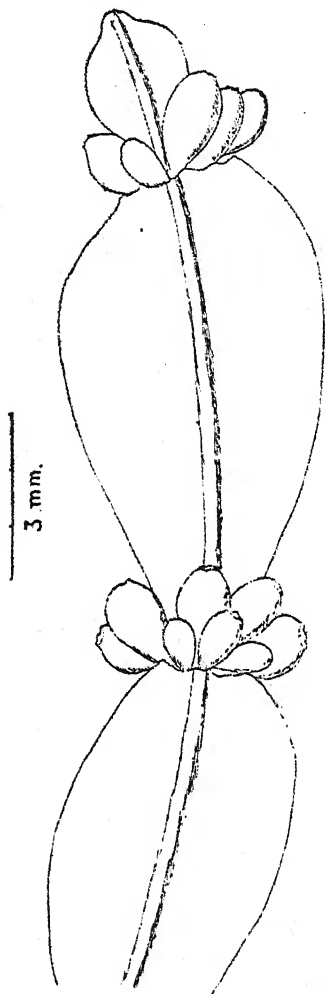
FIG. 1.

Fig. 1. A shoot bearing female flowers and " fruits "; the stem is flat and the internodes jointed, the shoot resembling a miniature *Opuntia*.

wall is sometimes thrown off as an empty sac although here the distal end is always ruptured. It would not be surprising to find a similar explosive mechanism in other Loranthaceae, especially in allied

species of *Viscum*, such as *V. articulatum*. The fact that no such cases have been recorded may be due to the scarcity of observations on the living plants.

It is always an interesting problem how certain species of plants, apparently having no very effective means of dispersal, have attained a wide geographical distribution. *Viscum japonicum* is a case in point. This plant occurs over an extensive area in the temperate Himalayas, and in the Khasia Hills, the Nilgiris,
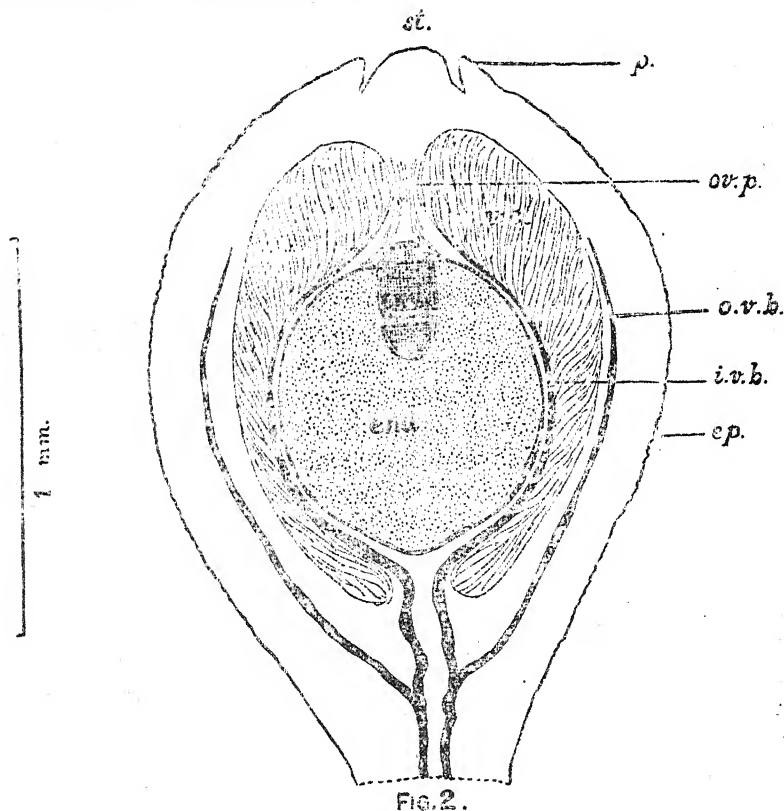
Fig. 2.

Fig. 2. A median longitudinal section of a nearly ripe "fruit". *st.* style, *p.* perianth, *ov. p.* remains of ovarian papilla, *o.v.b.*, *i. v. b.* outer and inner vascular bundles, *ep.* papillose epidermis with thick cuticle, *end.* endosperm (filled with starch), *emb.* embryo with projecting radicle, *v. c.* viscid cells with their proximal ends merging into the inner vascular bundles; the striation of the viscid layer as shown here roughly indicates the lie of the cells.

and the elevated parts of Ceylon. It extends into China, Japan and Australia; it has also been recorded from Mauritius and from the Sandwich Islands. As already stated, the fruit is unattractive and, so

far as I know, is never visited by birds. Apparently the only means of dispersal, therefore, is the explosive mechanism in the fruit possibly aided to some extent by the wind. It can only be by the merest coincidence that a seed thus carried through the air should get caught on the plumage of a bird. The chances of distant dispersal thus seem to depend more upon the small size of the seed than upon its viscid coat.

An equally interesting case is that of *Arceuthobium Oxycedri* which, like *Viscum japonicum*, has an explosive fruit, and a similarly wide distribution : Africa, Europe, North America, Himalaya and the Azores Islands. In discussing the distribution of this plant Guppy [7] expresses the opinion that birds probably actively disseminate the species, carrying the seeds adhering to their plumage. No one has, however, actually observed the seeds of this plant adhering to birds.

A detailed description of the floral and " fruit " structure (including the development) will be undertaken by Dr. P. Maheshwari but the main features of the ripe " fruit ", so far as they relate to the explosive mechanism, may be briefly described here. From the section shown in fig. 2 it will be apparent that there is a fundamental resemblance with *Arceuthobium* as described by Johnson and by Peirce. In a median longitudinal section the " fruit " of *V. japonicum* exhibits a deceptive resemblance with a *Cycas* ovule. There is a double set of vascular bundles ; the outer set runs in the perianth, while the inner is closely in contact with the inner ends of the viscid cells. These inner bundles are no doubt specially concerned with supplying the water which swells the viscid layer shortly before the ejection of the seed. A section cut from material which had been kept for several months in formalin-alcohol was placed on a slide in water ; it was noticed that the expansion of the viscid layer began at the thicker distal end, which became raised in the form of a dome pressing the apex of the fruit upwards. This pressure would gradually be communicated to the more proximal parts of the fruit, and as the lateral and basal parts of the viscid layer also expand, there would be an all-round compression of the seed which would only be relieved by an apical rupture, when the seed would be shot out like a bullet from a gun. In *Arceuthobium* both Johnson and Peirce figure the viscid tissue as a continuous dome-shaped cap to the seed [8]. If the sections figured by these authors are really median, *V. japonicum* differs from *Arceuthobium* in the fact that the viscid layer is perforated at the top. Another point of difference, already noticed above, is the absence, in *Viscum*, of any

[7] Guppy (1917), p. 427.

[8] Johnson, *loc. cit.* pl. X, fig. 9 ; Peirce, *loc. cit.*, pl. III, fig. 3.

well defined abscission layer. As in *Arceuthobium*, the radicle of the tiny ill-developed embryo projects from the starch-filled endosperm as a hemispherical knob which is capped by the remains ·of the ovarian papilla in the form of an inverted funnel, simulating the pollen-chamber of a cycadean ovule.

UNIVERSITY OF LUCKNOW,
   BOTANY DEPARTMENT.
   5th January, 1933.

Bibliography.

BAMBER, C. J. (1916).—Plants of the Punjab.

BENTHAM AND HOOKER (1880).—Genera Plantarum, vol. III, part (1).

BRANDIS, D. (1911).—Indian Trees.

COLLETT, H. (1902).—Flora Simlensis.

COOKE, T. (1906).—Flora of the Bombay Presidency, vol. II, pt. 3.

ENGLER IN ENGLER AND PRANTL. (1893).—Die Natürlichen Pflanzenfamilien, III Teil, Abt. 1. 1a, 1b.

FYSON, P. F. (1915).—Flora of the Nilgiri and Pulney Hilltops, vol. I.

GUPPY, H. B. (1917).—Plants, seeds and currents in the West Indies and Azores.

HAINES, H. H. (1921).—Botany of Bihar and Orissa, part II.

HOOKER, J. D. (1890).—Flora of British India, vol. V.

JOHNSON, T. (1888).—Ann. of Bot., vol. II, pp. 137–160.

KANJILAL, U. (1911).—Forest Flora.

KOORDERS (1912).—Exkursions-Flora v. Java, part II.

KURZ, S. (1877).—Forest Flora of British Burma, vol. II.

PARKER, R. N. (1918).—Forest Flora of the Punjab and Hazara.

PEIRCE, G. J. (1905).—The dissemination and germination of *Arceuthobium occidentale* Eng. Ann. of Botany, vol. XIX, pp. 99–113.

REINAUD DE FONVERT (1846).—"Note sur l' *Arceuthobium Oxycedri*." Ann. Sci. Nat., 3e Sér., T. VI.

STEWART, J. L. (1874).—Forest Flora of N. W. and Central India.

TRIMEN, H. (1895).—A handbook to the flora of Ceylon, part III.

A CONTRIBUTION TO OUR KNOWLEDGE OF THE ALGAL FLORA OF LAHORE SOILS

BY

HAR DYAL SINGH, M.Sc.,

Botany Department, Punjab University, Lahore.

Introduction.

In the present work an attempt has been made to describe those forms which grow in different types of soils found in Lahore and its vicinity. On account of the fact that only those algae have been described which could grow from the beginning of October, till the middle of May next, *i.e.,* mostly during the winter months, the number of forms studied has necessarily been a very limited one.

Method and Technique.

Samples of soils from different localities in Lahore representing gravel, sand, loam, and clay were collected. Gravel was collected from Shalamar, sand from Bahawalpur road, loam from gardens and fields and clay from Ravi bank and canal bank. Humus soil was collected from manure pits in Government College Botanic Garden and from the jungle on the left bank of river Ravi.

As a general rule composite samples were taken from a thorough mixture of at least 3 borings at 3 different places, to nearly the same depth in the same locality.

Most of the soil samples were alkaline in reaction, though a few were neutral.

Soil is collected after the whole apparatus and the culture media prepared for the cultivation and isolation of soil algae are sterilized. The culture media are sterilized in an autoclave at 1½ lbs. pressure for 15 minutes.

Into each of the sterilized flask a few grams of the soil to be examined is introduced by means of a sterilized spatula. The vessels are closed and placed under glass cases in the North window and left for some weeks to develop. The culture solution used for the purpose are the aqueous mineral salt solutions, a complete composition of which is given in the appendix. Evaporation from the surface of cultures takes place only slowly and if at all any water is needed sterilized distilled water is used.

Culture flasks were also set up in the manner of More and Karrer. Half an inch of burnt sand is placed in the conical flasks sterilised and plugged in the manner already described. A few grams of the soil to be investigated is introduced.

The culture solution is added before the introduction of the soil or a suspension of the soil in the mineral salt solution is added.

The sand in the flasks is slanted so as not to be wholly submerged, giving various moisture conditions.

According to the methods followed by Esmarch, a few petri-dishes were set up, but these did not yield any good result. Soil is packed in a petri-dish to a depth of 1 cm. well-moistened with sterile distilled water and the surface is covered with a piece of pure filter paper. These cultures are kept in diffuse light at a temperature of 20° to 25° C. The filter paper is moistened with sterile distilled water from time to time.

Growth in Culture.

The first sign of growth is observed after different intervals, in different soils, and even in the same soil, and in different periods of the year. On the surface first a scum gradually appears, which consists of bacteria. This scum next assumes a green tinge while at the same time on the soil surface many small tufts of green patches grow and become larger and larger every day. The whole culture medium assumes a light green colour owing to the presence of free floating unicells or filaments. The greenish scum also thickens gradually and the sides of the flask as well become covered with a green or dark green, gelatinous mass. As a result of continued growth, specially in culture-flasks in which Oscillatoria grows, the green mass becomes very sticky and difficult to be separated from the substratum. The culture-flasks which are kept moist show patches here and there of green velvety tufts which gradually increase in area. The soil becomes covered with a gelatinous mass and in it are entangled the filaments, etc. In flasks in which Diatoms develop the surface assumes a dull brown colour.

The first growth except of the uni-cellular algæ, when examined, reveals only a slight development of filaments very difficult to be identified. The abnormal condition of excessive moisture under which algæ grow in these cultures must tend to produce forms different from those that grow under natural conditions.

There is a good growth in culture flasks which are left undisturbed. Those which are frequently handled for observations or which are taken out of the glass house to be placed in direct sunlight, growth was very slow. The growth is impeded as it were, by

disturbance. It is also noted that the growth is quicker and more well-marked in culture flasks in which the culture medium-covers the soil completely, than in flasks which are kept simply moist and wet. Throughout November and December, there was no growth at all in culture flasks. The mean temperature of glass house during this time was 22° C. There was slight growth of Algæ in January and the first half of February when the mean temperature was 29° C. Growth was plentiful after mid-February. Up to this time the result seemed to be disappointing. Only six or seven species appeared. After mid-February the temperature ranged from 30° C. to 40° C.

It is also noted that growth is quicker in culture flasks when they are placed in the South window than when they are placed in the North window. In April, however, the flasks had to be placed in the North window as the direct rays in the South window proved very hot and impeded the growth. Four flasks were inoculated using the same soil and the same culture medium. Two of these were placed in the South window and two in the North. Growth was first observed in flasks which were placed in South window.

Different genera and even the different species are observed never to grow simultaneously in culture flasks having the same type of soil. In the floating scum various unicellular forms, for example, Chlorococcum, Protococcum, etc. appeared interspersed a little later with undeveloped filaments of Ostillatoria. Phormidium faveolarum and Nostoc filaments come out next and grow in almost all the flasks. Three weeks after that Anabaena and other species of Phormidium, Scytonema, Cylindrospermum sp. make their appearance in succession. Still later more species of Nostoc, Anabæna, Calothrix and Scytonema make their appearance. Next appear Hapalosiphon and Fischerella sp. After that long, broad, circular colonies of Nostoc appear which develop to form a large net-work of Nostoc filaments. At the same time some green algæ make their appearance at this stage, e.g., Ulothrix, Hormidium sp. etc. Diatoms appear quite early and continue to remain there for a considerable long time.

Isolation of Pure Cultures.

An attempt has also been made to isolate pure cultures of algae, by following Waksman (1) and Ward (6). Only in the former, pure colonies of Pleurococcus, Chlorococcus and Oscillatoria developed.

Cultures from Desiccated Soils.

A few soil samples were placed in sterilised papers (sterilized with absolute alcohol) for desiccation in the class room and left there for about 12 weeks. The soils were then inoculated in culture flasks

following Bristol (5). The following table indicates the locality from where the soil sample was obtained, the date on which it was placed for desiccation, the date on which culture flasks were set up and the date on which first growth was observed.

Locality		Date of desiccation	Date of inoculation	Date of first growth observed
Old garden	...	7–11–1931	13–3–1932	28–3–1932
Garden	...	9–11–1931	do.	26–3–1932
Cultivated field	...	15–11–1931	do.	26–3–1932
Uncultivated field	...	do.	do.	3–4–1932
Canal turf	...	do.	do.	25–3–1932
Grassy plot	...	15–11–1931	do.	30–3–1932

Growth has been found to be more plentiful in cultivated field soil than in any other type of soil. The number of species, however, is not the same as in fresh sample of soil. Oscillatoria, Anabaena, Phormidium, Nostoc and Diatoms only appeared in desiccated soils.

For the culture of diatoms, a modification of Miquel's medium (see Appendix) was used but not with very good results. The diatoms growing in this medium are not in any way larger in size than they ordinarily occur in the culture flasks and also the number of species does not show any increase in number.

A systematic list of the algæ recorded from 15 soil samples is given below. The classification followed is that of West and Fritsch (2).

Chlorophyceæ

Isokontae—Chaetophoraceæ.

Pleurococcus Naegelii (Chodat) from 4 samples.

Chlorococcum sp.—from 4 samples.

Ulotrichales.

Ulothrix sp. from 2 samples.

Hormidium sp. from 1 sample.

Cyanophyceæ

Chroococcales—Chroococcaceæ.

Aphanotheca naegelii, Wartm. from 3 samples.

Eucapsis minuta Fritsch. from 4 samples.

Aphanocapsa virescens (Hans) Rabh. from 2 samples.

Chroococcus minutus (Kutz) Nag. from 2 samples.

C. macrococcus (Kuetzing).

C. limneticus and

C. multicoloratus. from 3 samples.

Oscillatoria tenuis from 2 samples.

O. brevis and O. irrigua Kutz from 9 samples.

Isocystis sp. from 7 samples.

Phormidium foveolarum (Mont) Gom. from 7 samples.

P. unicinatum (Ag) Gam. and

P. ambiguum. from 2 samples.

Nostocaceæ

Nostoc verrucosum Linn.

N. ellipsosporum.

N. sphaericum.

N. pruniform.

N. spongiformæ from 7 samples.

Anabæna variabilis.

Anabæna sp. and

Anabæna inequalis (Kuetzing) from 7 samples.

Cylindrospermum majus from 2 samples.

Scytonmaceæ.

Scytonema sp. from 3 samples.

Rivulariaceæ.

Calothrix sp. from 3 samples.

Stigonemataceæ.

Hapalosyphon sp. from 2 samples.

Fischrella sp. from 1 sample.

DIATOMS—Bacillariaceæ.

1. Cyclotella Meneghiniana.
2. Synedra Ulna.
3. Navicula sp.
4. Navicula (Pinnularia) Gibbs
5. Navicula (Calonies) sp. ?

6. Gyrosigma scealproides, Rabh.

7. Amphora minutisima.

8. Cymbella sp.

9. Rhopalodia gibberula.

10. Nitzschia Kutzingiana.

11. Nitzschia Subtilis.

12. Nitzschia sp.

Summary.

Fifteen samples of Lahore Soils were collected and inoculated in culture flasks containing special enrichment culture media, favouring the growth and development of algæ. Four species of green algæ, 12 species of Diatoms and as many as 28 species of blue green algæ have been recorded.

Isolation of pure culture in Agar media was tried but only three species grew in the solid media.

A few soil samples were placed for desiccation for 12 weeks. Their algal growth was also studied, which was similar to that of the fresh soil forms.

LAHORE,
1st December, 1932.

Appendix.

Media used for the cultivation and isolating of algæ:—

1. Moor.

$NH_4 No_3$	...	0.5 gram.
$KH_2 PO_4$	...	0.2 ,,
$MgSo_4 7H_2o$	...	0.2 ,,
CaC_{l2}	...	.1 ,,
$FeSo_4$	...	Trace.
Distilled water	...	1,000 cc.

2. Bristol.

$NaNo_3$	...	0.5 gram.
$KH_2 PO_3$	...	.5 ,,
$MgSo_4$	...	0.15 ,,
CaC_{12}	...	.05 ,,
NaC_1	...	.05 ,,
FeC_{13}	...	.005 ,,
Distilled water	...	1,000 cc.

3. Detmer.

$Ca(NO_3)_2$	...	1.0 gram.
KH_2PO_4	...	0.25 ,,
KC_1	...	0.25 ,,
$MgSo_4\ 7H_2O$	...	0.25 ,,
Tap water	...	1,000 cc.

4. Pringshiem.

$NH_4\ Mg\ Po_4$	...	1.0 gram.
$K_2\ So_4$	...	0.25 ,,
$Fe_2\ (PC_4)_2$	...	Trace.
Water	...	1,000 cc.

5. Benecke.

$Ca\ (No_3)_2$	...	0.5 gram.
$Mg\ So_4\ 7H_2O$	...	0.1 ,,
$K_2\ HPO_4$	...	0.2 ,,
$Fe\ C_{13}$	...	Trace.
Water	...	1,000 cc.

6. Knops.

$Mg\ So_4$	...	0.25 grams.
$Ca\ (NO_3)_2$	...	1.0 ,,
$KH_2\ PO_4$	...	0.25 ,,
KC_1	...	0.12 ,,
$Fe\ C_{13}$	...	Trace.
Water	...	1,000 cc.
Strength =	...	0.172 per cent.

7. Klebs.

$Ca\ (NO_3)_2$	...	4 parts.
$KH_2\ PO_4$	...	1 part.
$Mg\ SO_4$	...	1 part.
KNO_3	...	1 part.

Made up in strengths from 0.2 to 1 per cent.

8. Miqual's medium for diatoms (modified).

Solution A 10.1 grams KNO_3 in 50 cc. of distilled water.

Solution B 2. grams $Na_2 HPO_4$ in 20 cc. water.

(1) c.c HCl concentrated 1 cc. $Fecl_2$.

(2) grams $CaCl_2$ dissolved in 20 cc water.

40 drops of A and 16 drops of B are added to 1,000 cc. of distilled water.

Bibliography.

1. WAKSMAN, S. A.—Principles of soil microbiology.

2. WEST, G. S. AND FRITSCH, F. E.—A treatise on the British Freshwaters Algae.

3. PASCHER, A.—Susswasser-Flora. Cyanophyceae Heft 12 and 5. Chlorophyceae.

4. TILDEN, J.—Minnesota algae.

5. BRISTOL, B. M.—On the Algal-flora of some desiccated English soils. Ann. Bot. 34, 35–79.

6. WARD, H. M.—Some methods for use in the culture of Algae. Ann. Bot. 13, 563–566.

7. BOYER, C. S.—Diatomaceæ of Philadelphia and vicinity, Philadelphia.

8. CHAMBERLAIN, C. J.—Methods in plant histology, Chicago.

9. SMITH, W.—Synopsis of the British Diatomaceæ, Vol. I and Vol. II.

10. WEST, G. S.—Algae, Cambridge, 1916.

11. WAKSMAN, S. A. AND FRED, E. B.—Laboratory manual of Gen. Microbiology, 1928.

ON THE MORPHOLOGY OF *RICCIA ROBUSTA*, KASHYAP.

BY

S. K. PANDE, M.Sc.,

Demonstrator in Botany, University of Lucknow.

(With 3 plates and 2 figures in the text).

Introduction.

Although the life-history of *Riccia* is fairly well known from a study of several foreign species (1, 2, 3, 7, 10, 11, 13), *R. sanguinea* is the only Indian species of which any detailed account is available (14). Prof. Kashyap (9) has described several new species from this country including *R. robusta*, but his account is mainly of taxonomic value. A casual examination of *R. robusta* growing here showed some variations from the Lahore plant and hence an investigation of this plant was undertaken.

The preliminary results of this study were communicated to the Indian Science Congress 1927 (15). In the following pages the author intends to give a somewhat fuller account of the species.

Occurrence.

R. robusta grows in moist and shady places during the cold weather. The plant is fairly common in the United Provinces and the Punjab and has also been recorded from Indore in Central India (9). Probably the species grows over the greater part of northern India. According to Prof. Kashyap (9) the plant may grow from 700 ft. in the Punjab plains to a height of 11,000 ft in Lahul and 13,000 ft. in Spiti. He observes that the specimens from Lahul are very small, have no tuberculate rhizoids and the spores are smaller than those of the type. Whether these differences are more than variations is not possible to decide in the present state of our knowledge of the Lahul plant.

Material and Methods.

Collections of the material were made from Lucknow and some of the neighbouring districts and the plants were fixed either in the field or after they had been brought to the laboratory and carefully washed to remove the adhering soil particles. Good fixation was obtained with medium chrom-acetic acid, Flemming's stronger solution and

acetic alcohol (3 parts absolute alcohol to 1 part glacial acetic acid). The material was thoroughly washed in running water in the case of aqueous fixatives, while absolute alcohol was used for the material fixed in acetic alcohol. Dehydration was begun with about 5 percent alcohol and the process was very gradual. Xylol was used for the most part as a clearing agent but in some cases cedar wood oil was also employed. Embedding was done in paraffin. Sections were cut from 4-10 microns thick and stained in safranin, and safranin and gentian violet.

Description.

Thallus.—*R. robusta* generally forms yellowish green spongy rosettes which may be incomplete in the case of the plants growing crowded together. Each thallus lobe has usually a broad median groove on the dorsal surface [see also Kashyap (9)].

The growth of the thallus takes place by means of an apical cell situated in the notch. This cuts off segments as in other species. The sexual organs and the assimilatory filaments are derived from the dorsal segments of the apical cell, while the scales and rhizoids are formed from the ventral segments (fig. 3).

Scales have not previously been noticed in this species (8, 9) ; but in his material the author has seen them near the growing points in many of the microtome preparations (figs. 1 and 3).[1] They are small, hyaline and rudimentary and dry up and fall off early. They are quite liable to be removed in cleaning the soil particles from the material for embedding in paraffin and hence will be observed only in very favourable preparations. It is not unlikely that in some cases the scales may not be formed at all. Both smooth-walled and tuberculate rhizoids are present.

Text-fig. 1 shows a transverse section through the growing point. It illustrates the dichotomous mode of branching. In fig. 2 is drawn another transverse section through an older part of the thallus. The air chambers are comparatively large and open to the exterior through indefinite pores.

---

[1] Mr. Nicholson in a recent note in "Annales Bryologici, Vol. V, p. 142, 1932" states that he has carefully examined *Riccia robusta* and in general appearance, the size of the spore and its sculpture he cannot find anything substantial to separate the plant from *Riccia crystallina* Goebel (Organography of Plants, English translation, Vol II, p. 28, 1905) states " In *Riccia crystallina*, which, according to Leitgeb, possesses no scales, I found them as very delicate structures, but perhaps there are some forms of this species where the scales are wanting, because an observer like Leitgeb would scarcely have overlooked them were they present.'—*Ed.*

R. robusta is monœcious. The sex organs are usually formed in acropetal succession. They arise from the young dorsal segments of the apical cells. The archegonial initial is almost spherical (fig. 9) while the antheridial initial is somewhat elongated (fig. 4).

Antheridium.—The antheridia were recorded for the first time by the writer in 1927 (15). Previously their presence was regarded doubtful as may be seen from the following extract quoted from an earlier contribution of Prof. Kashyap (8) :—" The presence of antheridia is doubtful. A large number of plants were dissected but no papilla[e] were ever observed and only once structures which looked like antheridia were seen. The possibility of parthenogenesis should be kept in mind." In a later publication (9) he says, however, that the plant is monœcious and the papillæ " not distinct." The development is of the usual type and is illustrated in figs. 4–8.
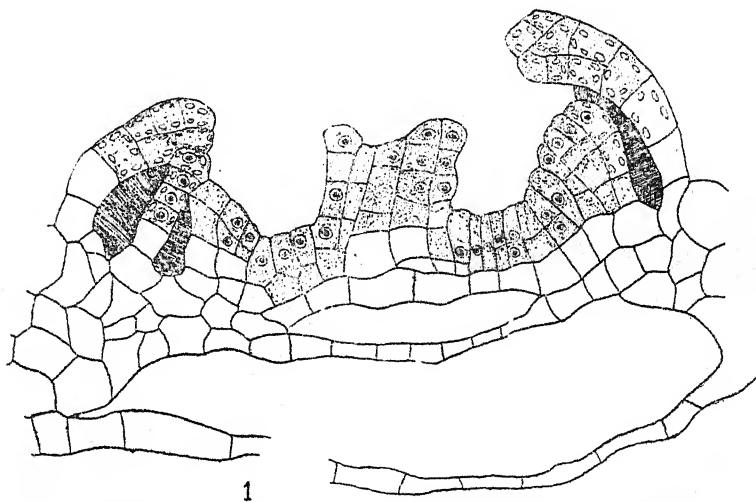
Fig. 1. T. S. of thallus through growing point.   × 315.

Archegonium :—(*a*) *Normal structure.* The development of the archegonium is in general like that of the other species of *Riccia*. Different stages are shown in figs. 9–13. The mature archegonium has four neck canal cells (fig. 13). This appears to be the usual number in *Riccia* so far as known.

(*b*) *Abnormal archegonium.* Figs. 16 and 17 are drawn from two sections of the venter of a mature archegonium. Both the ventral and the neck-canal cells have disorganized. The venter is occupied

by two egg-like bodies.   On comparing this with a mature unfertilized
egg (fig. 14) and a fertilized egg (fig. 15) one is inclined to think that
this is probably an abnormal archegonium with two eggs.
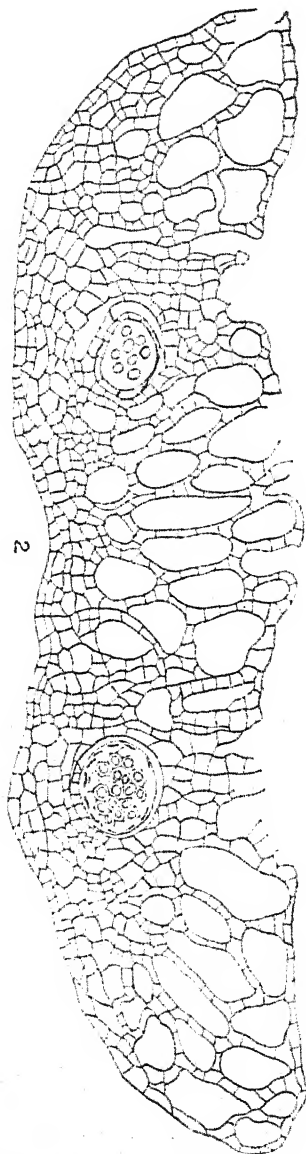
Fig. 2. T. S. of thallus through older part.   × 45.

(c) Abnormal archegonial chamber.—An interesting feature observed is that sometimes two archegonia occur in one chamber (Figs. 18–20). Both of these may produce sporogonia. Such a condition is rather unusual for *Riccia* and reminds us of *Corsinia* where archegonia are produced in sunken groups on the dorsal side of the thallus. In *R. pathankotensis* according to Prof. Kashyap (9) sporogonia are found in groups and sometimes as many as six of these occur together touching each other without any intervening green tissue. He, however, remarks that the absence of the vegetative tissue is due to disorganization of the small amount of tissue present in the beginning. From what has been seen in *R. robusta* it appears not unlikely that in some cases at least, these sporogonia in *R. pathanko-tensis* might arise from a group of archegonia lying in the same chamber. A further investigation is desirable to ascertain this point.

Fertilization.—As the egg matures the ventral and the neck-canal cells disorganize and produce a mucilaginous substance. Fig. 21 shows the male nucleus in contact with the female nucleus. The former is about 1/2 the size of the latter. The egg cytoplasm is much vacuolated, the nucleus is enlarged and the chromatin is collected in the form of short rods. Following the act of fertilization an enlargement of the calyptra takes place and often it becomes two-layered even before the initiation of division in the oospore (fig. 15), but sometimes it remains only one cell thick even at the time of the first division (fig. 22).

Development of sporophyte.—The zygote divides by an oblique wall (fig. 22). A review of the literature on *Riccia* shows that this wall may be either oblique or horizontal and sometimes both the conditions may be met with even in the same species. In the accompanying table are recorded the observations of different authors regarding the position of this wall in several species.

The second wall is more or less at right angles to the first resulting in a quadrant (fig. 23). This is followed by a third wall which meets the other two at right angles and produces the octant stage (fig. 24). In the embryogeny of *R. natans* both Garber (7) and Lewis (11) noticed that sometimes walls parallel to the first are laid in the epibasal and hypobasal cells and produce a filamentous embryo of superposed cells. Black (2) also observed a filamentous embryo in some cases in *R. Frostii*. Evidently, as Fagan (13) remarks, both the filamentous and the octant types occur in the genus *Riccia*, but the latter is more frequent. In fact, the author is not aware of a single species where the filamentous type of embryo alone is formed, whereas in several species only the octant type occurs. The

cells of the sporophyte now divide without any regular sequence and give rise to a more or less spherical mass of cells. The wall is cut off from the outermost cells by periclinal divisions (fig. 25). It remains one cell thick throughout.

Author.		Species.	Direction of the first division wall.
Garber 1904 (7)	...	*R. natans*	generally transverse, may be oblique.
Lewis 1906 (11)	...	*R. natans*	generally transverse, may be oblique.
Beer 1906 (1)	...	*R. glauca*	usually obliquely transverse.
Black 1913 (2)	...	*R. Frostii*	usually oblique.
Pande 1924 (14)	...	*R. sanguinea*	oblique.
Campbell 1929 (3)	...	*R. glauca*	more or less inclined to the axis of the archegonium, approaching usually the horizontal.
Pagan 1932 (13)	...	*R. crystallina*	transverse or inclined to the major axis of the archegonium.

With the increase in the size of the sporogonium an enlargement of the calyptra and venter takes place. After the last division the archesporial cells begin to round off (fig. 26). A little later disorganization of the walls separating the spore mother cells commences and by the time of the heterotypic mitosis the latter lie free in the sporogonial cavity. Garber (7) and Lewis (11) have both observed a large amount of nutritive material around the spore mother cells of *R. natans*. This food material is derived from the surrounding tissue. Beer (1) did not find any such material in *R. glauca*. Black (2) noticed a mucilaginous substance which has great affinity for stains like gentian violet and Bismarck brown in *R. Frostii*. Quite recently Pagan (13) who described food material around the spore mother cells of *R. crystallina*, concluded that the food material might have come from four different sources; viz., "storage products of photosynthesis within the cells of the gametophyte, disorganization of the cells of the

sporangium wall and calyptra, decomposition products of the cell walls between the spore mother cells, and disintegration of some of the spore-mother cells themselves."

In *R. robusta* a mucilaginous substance which stains with gentian violet has also been observed. It appears to be derived in the first place from the decomposition of the original walls of the spore mother cells. Later on the decomposition products of the sporogonial wall and the calyptra are also added on to this material.

The wall of the sporogonium (amphithecium) may break down early or late. Thus according to Black (2) in *R. Frostii* it gets partly disintegrated by the time the spore mother cells are formed ; while Garber (7), confirmed by Lewis (11), remarks that in the case of *R. natans* the wall could be distinguished even when the spores were almost ripe. In *R. crystallina* Pagan (13) noticed that the wall breaks down early in some cases while in other cases it remains intact till the time of the reduction division. He regards this as a question of food supply and correlates the late disappearance of the wall with the failure of some of the potentially sporogenous tissue to produce spores.

The first signs of the disintegration of the sporogonial wall in *R. robusta* are seen at the time of the heterotypic division and the process is completed by the time the spores have reached the stage shown in fig. 41. It is interesting to note that though all the arche-sporial cells in *R. robusta* produce spores and there are no nourishing cells, the wall breaks down quite late. The absorption of the inner layer of calyptra starts a little later than that of the wall and the process is completed in a longer time. The outer layer of the calyptra contains abundant starch (fig. 27) and breaks down much later.

Sporogenesis.—Farmer's paper of 1893 (4) was the earliest cytologi-cal contribution on sporogenesis in the Hepaticæ. This was followed by another paper dealing with *Pallavicinia decipiens* (5). In 1895 (6) he published a more comprehensive account describing the process in several liverworts. His researches stimulated the study of the subject and since then several papers have appeared dealing with this important phase of the life-history in many liverworts.

In the course of this paper reference will be made to some of these.

Garber (7) working on *R. natans* observed prominent asters in the sporophyte cells but no centrosomes were seen.

Lewis (11) records no nucleolus in the spore mother cells of *R. natans* and *R. crystallina*. He records the synaptic stage at which the chromatin is collected eccentrically in an irregular mass. Neither a centrosphere nor a centrosome was seen.

Beer (1) studied sporogenesis in *R. glauca* and observed a definite and large nucleolus. Sometimes the latter was composed of several deeply staining granules embedded in a faintly staining matrix.

Black (2) has recorded scanty chromatin and small spindles in *R. Frostii*. A definite nucleolus was observed but no centrosphere or centrosome was seen. In the heterotypic division a granular and distinct but incomplete cell plate is formed. The homotypic division is simultaneous and the two spindles may be in the same plane or at right angles to each other.

The spore mother cells of *R. robusta* are not very favorable for the study of sporogenesis. A resting spore mother cell is shown in fig. 28. The nucleus is large and the membrane is distended. It contains a prominent nucleolus which in some cases is broken up into two or three coarse and prominent granules (fig. 29). Sometimes the nucleolus is composed of several granules embedded in a matrix (fig. 30) recalling the condition described by Beer (1) in *R. glauca*.

Gradually the protoplast rounds itself off and becomes separated from the primary wall, leaving a clear space between itself and the membrane. In some cases this space is occupied by a number of strands of granular material (fig. 31). A somewhat similar condition is described by Beer (1) in *R. glauca*, but he obtained cellulose and pectose reactions with these strands. He thinks these strands to have been derived from the secondary thickening layer laid down around the protoplasts. Leitgeb, as quoted by Beer (1), believed that the space between the protoplasts and the primary membrane was occupied by a homogeneous mucilage and there were strands of material running between the primary wall and the protoplasts. He believed these strands to represent food material diffusing in from the outside. The strands in *R. robusta* are composed of a granular material without any obvious difference from the contents of the spore mother cells and this leaves little doubt regarding their identity with food material.

At the time of the heterotypic mitosis the mother cells lie free in the sporogonial cavity, the nucleus is somewhat elongated and the cytoplasm is granular. As the division stage draws near the nucleus frequently presents the appearance drawn in fig. 32. This undoutedly represents synizesis. Several stages in both the heterotypic and homotypic mitosis have been obtained, but the number of chromosomes could not be counted. In fig. 33 the chromosomes are seen at the equator. A later stage is shown in fig. 34. The spindle fibres are quite distinct. Some of them extend from pole to pole, while others end at the equator or diverge in the protoplasm. The telophase stage is drawn in fig. 35. The cell plate has been laid down.

There seems to be a very short period of interkinesis. The homo-typic division is simultaneous and the spindles may be parallel (fig. 36) or at right angles to each other (fig. 37). Fig. 37 shows the late telophase in the homotypic division. The cell plate from the first mitosis has become more pronounced and the initial stage is seen in the formation of the other cell plate. By further growth of the cell plates which is centrifugal the division of the protoplast into four is brought about (fig. 38). All the spore mother cells give rise to spores. Quite recently Pagan 13) has observed abortive (nutritive) cells in *R. crystallina* which arise from a diversion of the sporogenous tissue. These cells may become abortive before they round off or before the reduction division and in some cases when the division of the spore mother cells has taken place. He regards these sterile cells as the fore-runners of the elaters of the higher forms.

The development of the spore coats agrees with the account given by Black (2) for *R. Frostii*. Inside the primary wall of the spore mother cell another layer of mucilaginous nature is formed (fig. 39). Within this, a second layer which projects into the first at numerous places is developed (fig. 40). This second layer ultimately becomes the rough outer spore coat (figs. 41 and 42). Finally the endospore is formed. The exospore is yellow in young condition, becoming orange as the spores grow older and ultimately black. The surface sculptur-ing of the spore is shown in fig. 43. As has already been noted by Kashyap there are 6 or 7 reticulations across a diameter in the spores of the Lucknow plant, and the wing is either narrow or almost absent. The spores in the Lahore plant have only 4 or 5 reticulations and the wing is generally broad (9). The size of the spores is the same in the two cases.

R. robusta shows a striking resemblance to *R. crystallina* (as may be inferred on comparison with the figs. and description given by Macvicar (12). Nicholson (12a) observes that he has examined the species very carefully but found no substantial difference in the general appearance and the size and sculpture of the spores to separate it from *R. crystallina*. Pagan (13) has, however, described sterile (nutritive) cells in the latter species, but no such cells were seen by me in the case of *R. robusta*.

DEPARTMENT OF BOTANY,
UNIVERSITY OF LUCKNOW,
    15th November, 1932.

Summary.

1. On the whole the observations of Prof. Kashyap regarding the vegetative characters of the plant have been confirmed except with regard to the scales which have been described in *Riccia robusta* for the first time by the present author.

2. The species is monoecious. Definite antheridia were recorded for the first time by the writer in 1927. The development of these is of the usual type.

3. The mature archegonium has four neck canal cells.

4. Abnormal archegonia with two egg-like bodies have been described.

5. Normally the archegonia occur singly in their own cavities, but occasionally two were noticed inside a common chamber.

6. The first division wall in the oospore is oblique.

7. An octant stage was always seen.

8. The spore mother cell has a definite nucleolus.

9. The two spindles in the homotypic mitosis may lie parallel or at right angles to each other.

10. No centrosome or centrosphere was seen.

11. The outer spore coat has the sculpturing in the form of reticulations.

12. The spores in the Lucknow form have a narrow wing or none and the number of reticulations is 6 or 7.

Literature.

1. BEER, R.—On the development of the Spores of *Riccia glauca*. Ann. Bot. 20: 275-291. 1906.

2. BLACK, CAROLINE A.—The Morphology of *Riccia Frostii* Aust. Ann. Bot. 27: 511-532. 1913.

3. CAMPBELL, D. H.—The Structure and Development of Mosses and Ferns. 1928.

4. FARMER, J. B.—On the relation of the nucleus to spore formation in certain Liverworts. Proc. Roy. Soc., London. 54: 478-480, 1893.

5. „ „ Studies in Hepaticae: On *Pallavicinia decipiens* Mitten. Ann. Bot. 8: 35-52. 1894.

6. „ „ On Spore formation and Nuclear division in the Hepaticae. Ann. Bot. 9: 469-523. 1895.

7. GARBER, J. F.—The life-history of *Ricciocarpus natans*. Bot. Gaz. 37: 161-176. 1904.

8. KASHYAP, S. R.—Liverworts of the Western Himalayas and the Punjab, with notes on known species and descriptions of new species. Journ. Bomb. Nat. His. Soc. 343–352. Jan. 1916.

9.   „   „   Liverworts of the Western Himalayas and the Punjab Plain, Part 1. 1929.

10. LEITGEB, H.—Untersuchungen über die Lebermoose. IV Die. Riccieen 1879.

11. LEWIS, C. E.—The embryology and development of *Riccia lutescens* and *Riccia crystallina*. Bot. Gaz. 41 : 109–138. 1906.

12. MACVICAR, S. M.—The Student's Hand-book of British Hepatics. 1926.

12 a. NICHOLSON, W. E.—Annales Bryologici, V : p. 142. 1932.

13. PAGAN, F. M.—Morphology of the sporophyte of *Riccia crystallina*. Bot. Gaz. 93 : 71–84. 1932.

14. PANDE, S. K.—Notes on the Morphology and Biology of *Riccia sanguinea* Kash. Jour. Ind. Bot. Soc. IV : 117–128. 1924.

15.   „   —Notes on *Riccia robusta* Kash. Proc. Ind. Sc. Congress, Lahore, 1927.

Explanation of Plates I-III.

PLATE I.

Fig. 3. L. S. Through growing point of thallus *ap. c.*, apical cell ; *sc.* scales.   × 343.

Fig. 4. Antheridial initial. × 525.

Figs. 5–7. Stages illustrating the development of antheridium. × 515.

Fig. 8. Mature antheridium, somewhat diagrammatic. × 343.

Fig. 9. Archegonial initial. × 515.

Figs. 10–13. Different stages in the development of archegonium, *vc.*, ventral canal cell ; *nc.*, neck canal cell ; *e.*, egg. × 515.

Fig. 14. L. S. of mature unfertilized archegonium. × 343.

Fig. 15. L. S. of fertilized archegonium. × 343.

Figs. 16–17. Two successive sections of an abnormal archegonium. *e.*, egg. × 343.

PLATE II.

Figs. 18–20. Serial sections through an archegonial chamber containing two archegonia. × 343.

Fig. 21. Fusion of the male nucleus with the egg nucleus. × 515.

Fig. 22. Oospore dividing. × 515.

18

20

calyp.
W.

27

19

25

22

♂
♀
21

23

24

26
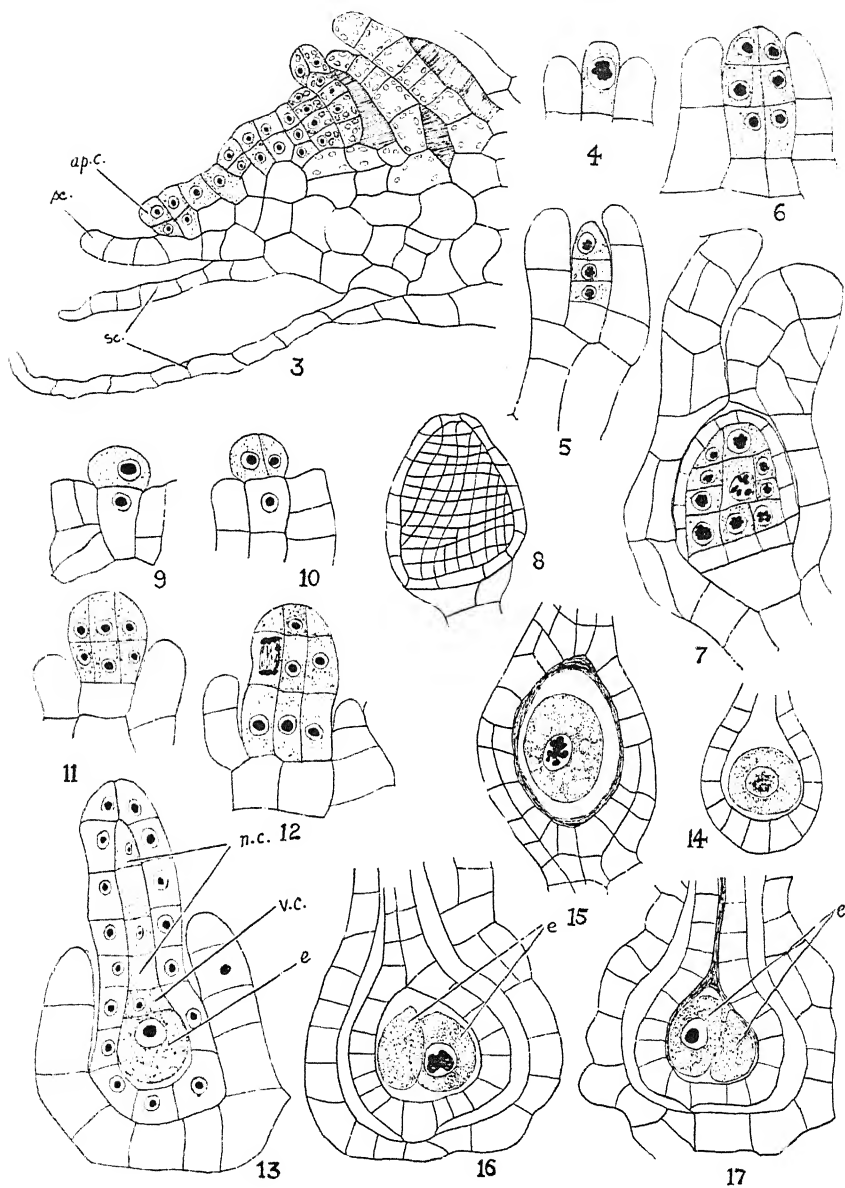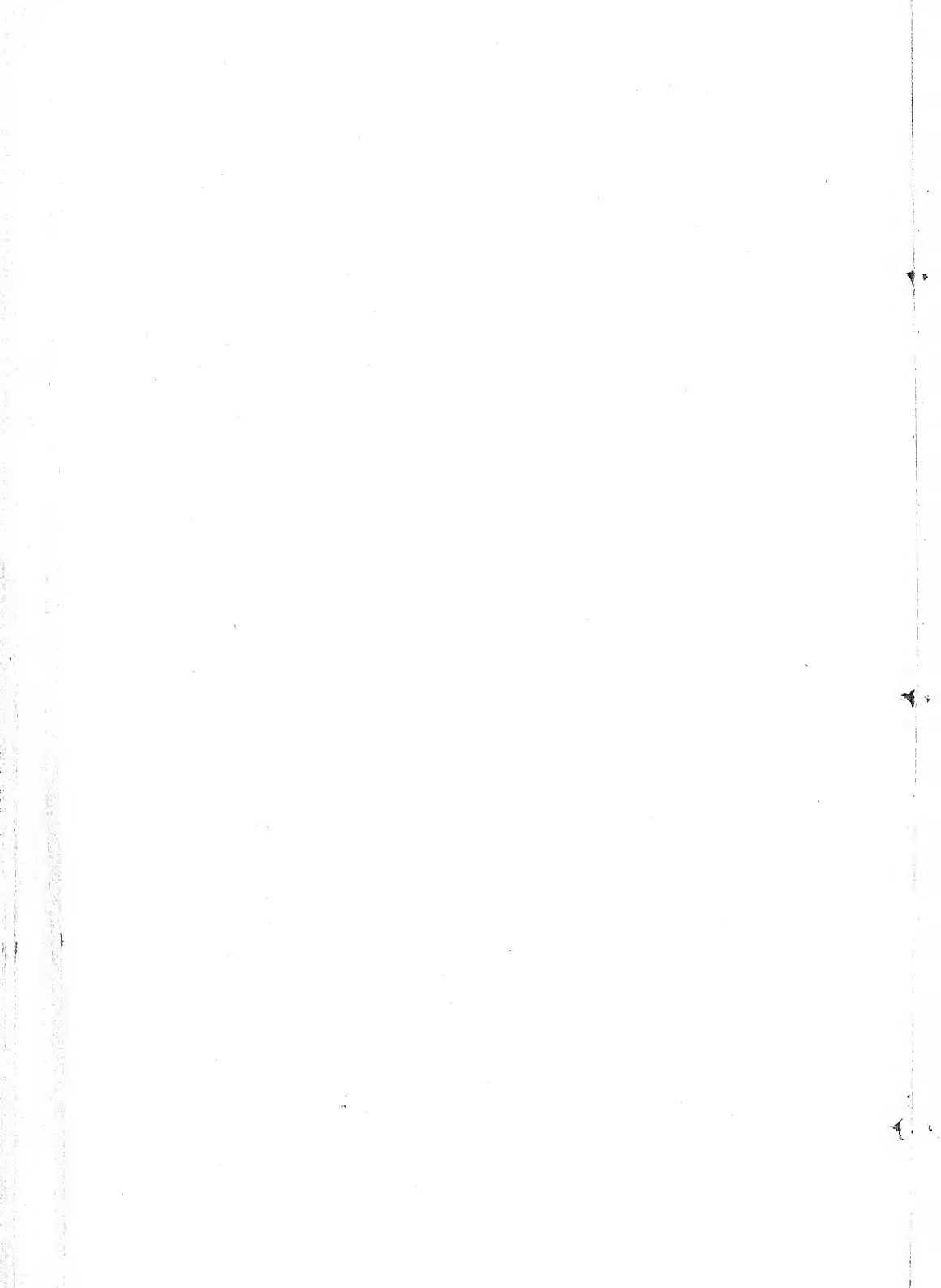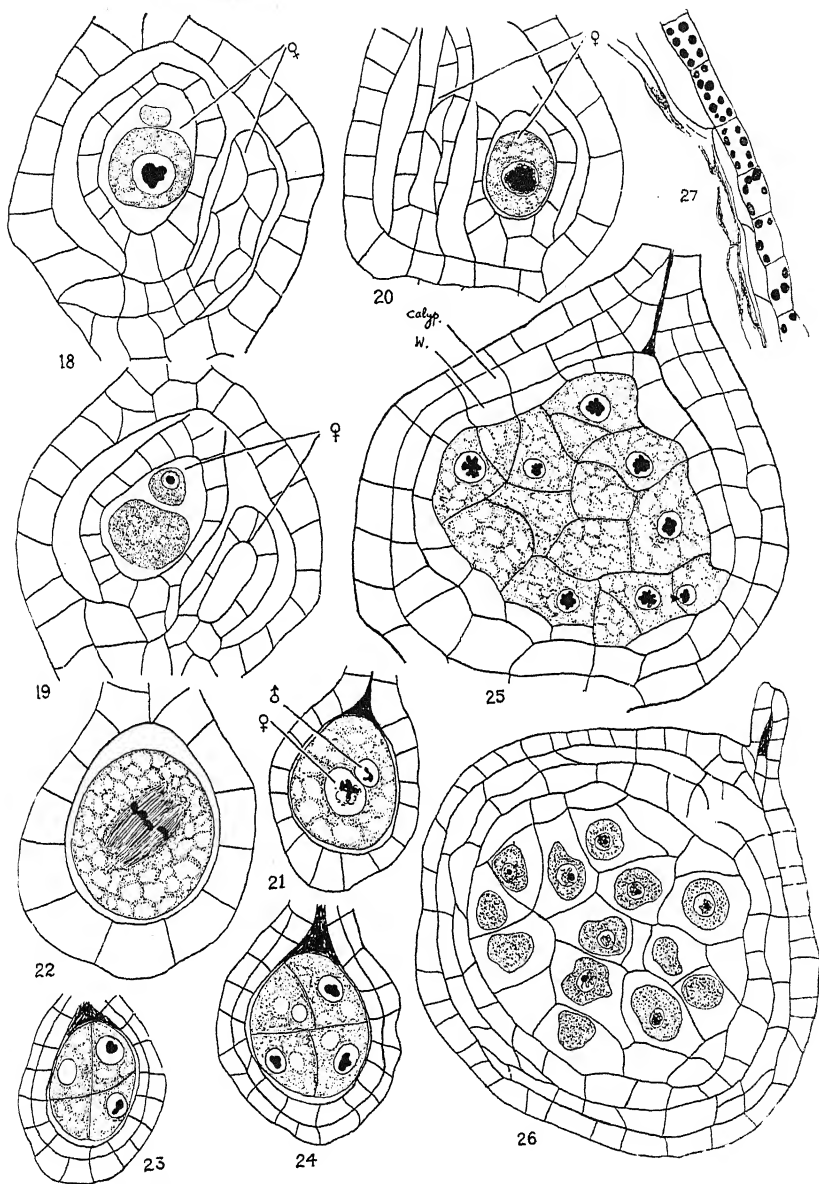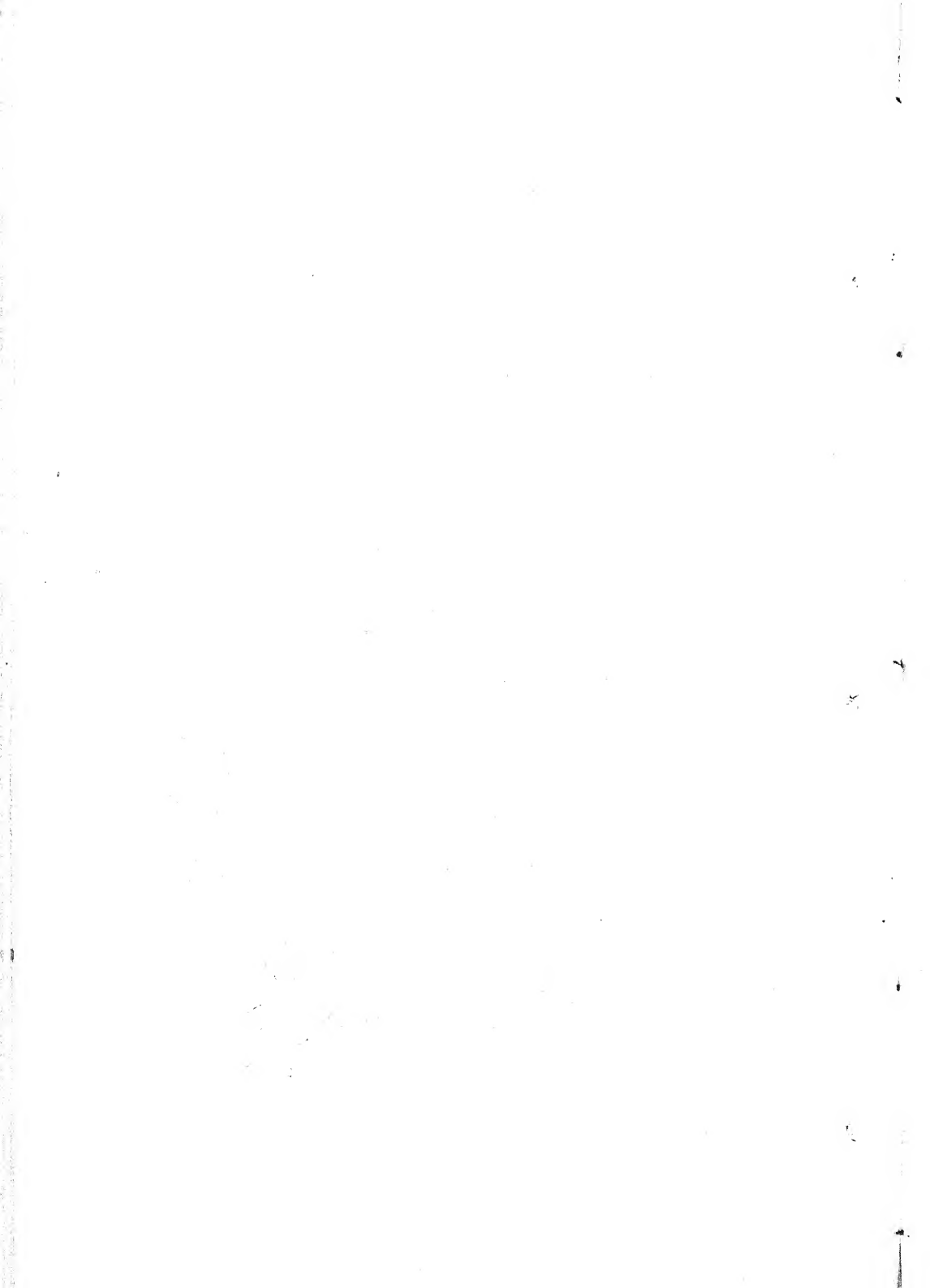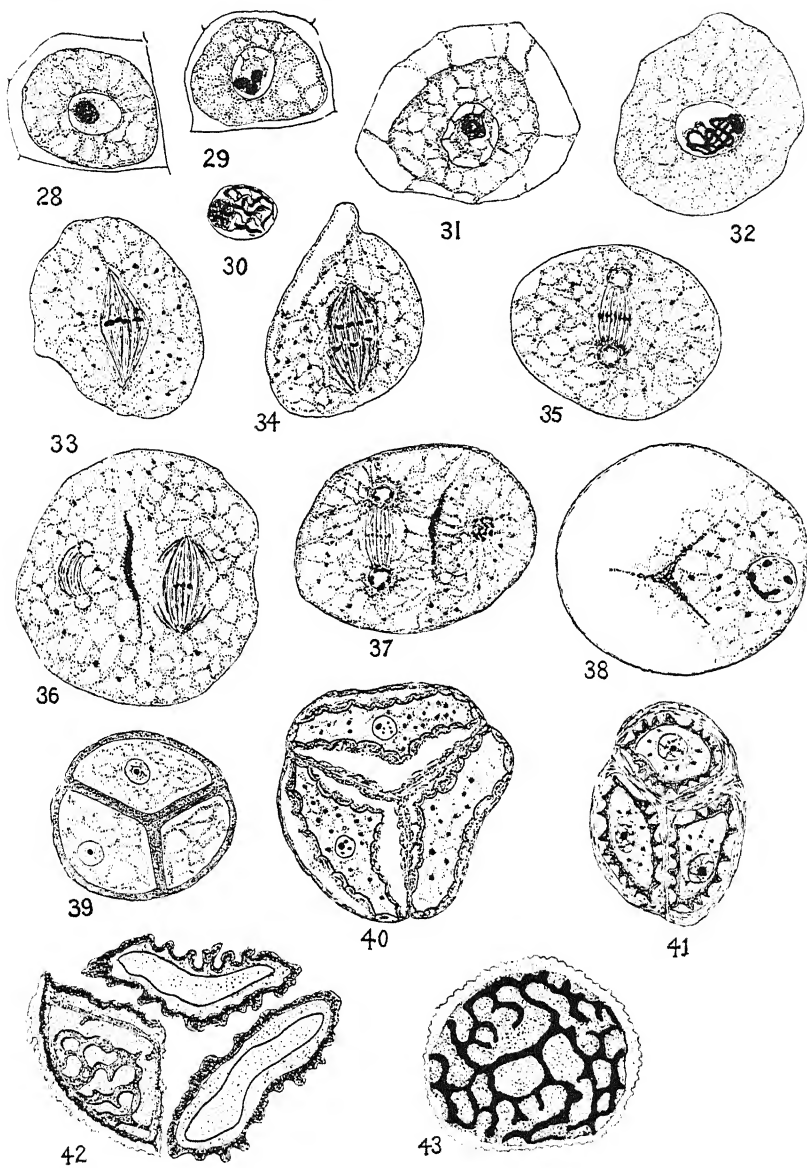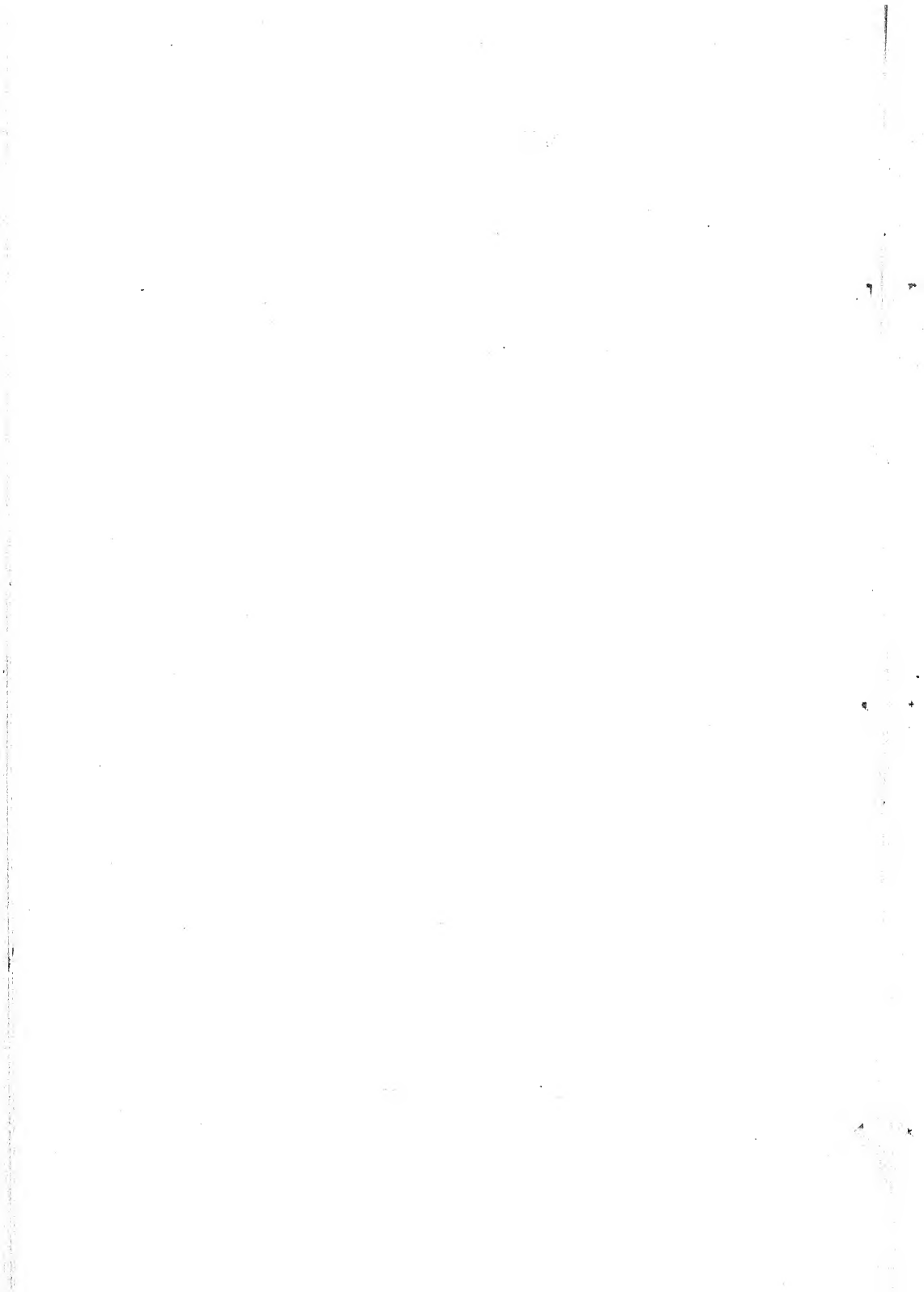
Fig. 23. Four celled embryo. × 343.

Fig. 24. Embryo at the octant stage. × 343.

Fig. 25. Young sporophyte showing the wall formation. *w.*, wall. *calyp.*, calyptra. × 343.

Fig. 26. Older sporophyte. × 152.

Fig. 27. A portion of the calyptra and the sporogonial wall from a sporophyte at the time of the first division of the spore mother cells. Note abundant starch in outer layer of calyptra. × 343.

PLATE III.

Figs. 28–29. Spore mother cells at the resting stage. × 430.

Fig. 30. Nucleus of a spore mother cell showing compound nature of nucleolus. × 730.

Fig. 31. Spore mother cell. Note the strands connecting the protoplast with the primary wall. × 570.

Fig. 32. Spore mother cell showing synizesis. × 730.

Fig. 33. Heterotypic mitosis with the chomosomes at the equator. × 580.

Fig. 34. Later stage in heterotypic mitosis. × 580.

Fig. 35. Telophase in heterotypic mitosis. × 580.

Fig. 36. Homotypic mitosis. The two spindles are parallel to each other. × 734.

Fig. 37. Homotypic mitosis. The two spindles are at right angles to each other. × 580.

Fig. 38. Late telophase in homotypic mitosis showing organization of nuclei and extension of plasma membranes. × 734.

Fig. 39. Young spore tetrad. × 405.

Fig. 40. Older spore tetrad showing the formation of second spore membrane. × 343.

Fig. 41. Older spore tetrad. × 343.

Fig. 42. Spore tetrad mature. × 343.

Fig. 43. Spore in surface view showing the sculpturing. × 330.

ON THE STRUCTURE OF A NEW SPECIES OF IND AN MOSSES *PHYSCOMITRELLOPSIS INDICA,* Dixon, sp. nov. FROM BENARES.

BY

K. M. GUPTA, M.Sc. (LUCKNOW),

Research Fellow, Botany Department, University of Lucknow.

Introduction.

Professor B. Sahni kindly placed at the author's disposal some material of a moss fixed in chrom-acetic acid and preserved in 90% alcohol. It consisted of vegetative and fertile shoots together with sporogonia at various stages of development except the ripe ones. A few specimens sent for determination to Mr. H. N. Dixon were identified as a new species of *Physcomitrellopsis* Broth. and Wager, a South African genus first described by Dixon in 1922 [1]. The only species so far known is *P. africana* Broth. and Wager. The plant now described represents the first record of this genus in India [2], and although essentially similar to the African moss, it is sufficiently distinct to be regarded as a new species.

Description.

Diagnosis:—"Dense gregaria; mollis. Caules 3–4 mm. alti parce ramosi. Folia superiora circa 3 mm. longa, e basi elongata, angusta, lanceolata vel spathulato-lanceolata, longe acuminata, parte dimidia superiore argute, sat distanter denticulata. Costa tenuis, supercurrens, ariolatio laxa, cellulae superiores 100–$200\,\mu$ longae, 30–$40\,\mu$ latae, parietibus tenuibus; marginales seape angustiores, inferiores majores, praelongae."

"Theca pedunculo circa ·60 mm. longo, *sphaerica* vel *oblate sphaeroidea,* brivissime obtuse apiculata, leptodermica, maturitate, *irregulariter* dehiscens. Exothecii cellulae laxae, hexagonae, homogeneae nisi propter lineam horizontalem e parietibus incrassa-

---

1. Dixon (1922), p. 107.

2 I have failed to find any record of this genus in India in the literature published after 1922 which I have consulted. See bibliogia 1 y

tis instructam, orificium simulantem. Calyptra magna, mitriformis, basi *profunde laciniata.* Spori 30–40µ, teneriter muriculata."

Besides the characters mentioned in the above diagnosis kindly supplied by Mr. Dixon for the Indian species, I may add that a few stomata are also found on the apophysis of the capsule.

Habit:—Dense, gregarious, soft and tufted with deep green colour, 3–5 mm. high, usually unbranched, sometimes sparingly branched (Fig. 1).

Habitat:—Damp ground under a bridge, Benares (Professor B. Sahni. October 1919). Recently, however, the same species has been found to be common in Lucknow. Further examination of the mosses might prove it to be a common species occurring in the plains of northern India.

Stem and *rhizoids:*—The stem is small and soft, becoming slightly brownish at the base. The internal structure is rather simple, with no differentiation into hadrome and leptome, but the thin-walled cells of which it is composed are smaller in the middle and bigger towards the periphery (Fig. 4). At the base of the stem are attached numerous multicellular rhizoids with oblique end-walls.

Leaf:—The leaves are spirally and densely disposed. They are distantly serrate (Fig. 2) with slight variation in the length of the perichaetial and the lower leaves. They are about 3 mm. long, narrowly lanceolate to spathulate-lanceolate with the midrib extending up to the tip in some cases. Upper cells are 100–200µ long and 30–40µ broad. The marginal cells are narrower and the serrations on the leaf are caused by the individual cells (Fig. 3). The leaf is a single layer of cells thick and is about 20–30 cells broad. The midrib is about 10 cells in thickness; of these two or three central cells (as seen in the cross-section) (Fig. 8) are specially narrow and thick-walled, simulating a rudimentary conducting strand. These thick-walled cells do not pass downwards into the stem.

Sex organs:—The disposition of the sex organs is always at the stem apex which is completely surrounded by the perichaetial leaves. The Indian species is distinctly monœcious and synœcious, that is, it bears antheridia and archegonia on one and the same head. Intermixed with the sex organs are found two kinds of paraphyses (Fig. 29). Fig. 8 shows the antheridial group segregated from the archegonial group on the fertile top, though

as a rule the two sexes are mixed together. As several antheridia and archegonia are borne on the same head, it is unlikely that all the sex organs in the head arise from a single apical cell.

(a) *Antheridium:*—The antheridium is a small, sessile, more or less ellipsoid body with the base slightly narrowed. The wall is one layer thick with a few unusually large cells at the top and at the base (Figs. 11, 22–24). The central bigger cell of the top region seems to disorganise when the antheridium ripens and the spermatozoids escape (Figs. 22–24); while the lower cells remain embedded in the stem apex. The spermatozoids are small coiled structures thickened at one end (Fig. 22). The cilia could not be made out.

(b) *Archegonium:*—The details in the development of the archegonium have not been followed but from what has been observed, it seems to have the usual structure (Figs. 9, 10). The pedicel is massive and remains embedded in the stem apex. The number of neck canal cells is usually four (Fig. 10). The size of the ventral canal nucleus is practically the same as that of the egg nucleus. No cell wall has been observed between these two nuclei, a fact which may indicate that this wall dissolves soon after the division of the egg nucleus.

(c) *Paraphyses:*—These are of two kinds, both kinds are four to five cells in length, the topmost cell becoming club-shaped in the one and capitate in the other. The terminal cell in the former has a prominent nucleus and dense contents, while in the latter it has dark green chloroplasts besides, all arranged along the periphery (Figs. 12, 13).

Protonema:—No protonema could be detected in the preserved material from Benares; but the examination of the fresh sample from Lucknow revealed that it has the structure of a branched filament. It usually arises from the rhizoids under suitable conditions.

Sporophyte. (a) *Foot* and *seta* :—The foot is about 0·6 mm. long and is deeply immersed in the stem apex which closely envelops it (figs. 21, 31). The seta is quite short.

(b) *Apophysis:*—The apophysis of the capsule is comparatively small and bears a row of stomata with their slits placed transversely or longitudinally (Fig. 7). The stoma is somewhat sunken below the general surface of the capsule and is surrounded by one big annular guard cell (Fig. 6) as in *Funaria*.

(*c*). *Capsule:*—The ripe capsule, either quite spherical or slightly flattened vertically, is about 1·0 mm. long and 0·7 mm broad. It has a small rounded apiculum (Figs. 29, 31). The capsule, at about 2/3 of its height, is divided into two well defined portions by a horizontal row of cells. The cells of this row are comparatively smaller and denser in contents than the adjacent cells on the wall of the capsule and have their lower cell wall thickened (Fig. 5). So far as observed the capsule breaks along this line of dehiscence; and the upper part of the capsule, with regularly arranged cells, is pushed aside like an operculum, though there is no distinct annulus (Fig. 31). The lower part of the sporogonium with thin-walled outer cells encloses the spore sac.

There is a definite columella which is gradually absorbed as the capsule ripens (Fig. 17). A well defined annular air space surrounds the archesporium (Figs. 14–16). The big and deeply lobed mitriform calyptra, best seen covering the younger sporogonium completely, splits at the base in many places and gradually shrivels away with the maturity of the capsule (Figs. 25–28). The spores are 30–40μ in diameter and have a large number of minute projections over them (Fig. 30).

The sporophyte develops by means of two apical cells, upper and lower (Fig. 18). The capsule during development gradually assumes a spherical shape, the shape of the young capsule being elliptic.

Theoretical.

Brotherus and Wager founded the genus *Physcomitrellopsis* on material which was essentially similar to *Physcomitrella* except in the shape of the calyptra and the extent to which it covers the young sporogonium. It is to be doubted whether we are justified in separating genera on such single characters as the one mentioned above; because a detailed comparative study of the development of the vegetative and the reproductive organs might prove them to be generically identical.

The figures given by Mr. Dixon in his brief description of the African species of *Physcomitrellopsis* are too few and incomplete, but as far as ascertained, the Indian species seems to be quite distinct in having an oblate spherical capsule with a distinct line of dehiscence, in contrast to the elliptic shape of the capsule with no indication of a line of dehiscence in the African form. The occurrence of two closely allied species of the same genus in such widely

separated lands (India and Africa) is a fact of some interest. The distinguishing features of the two species are as follows:—

Physcomitrellopsis indica, Dixon	P. africana, Broth. and Wag.
1. Capsule oblate spherical in adult condition, with a distinct line of dehiscence.	1. Capsule elliptical, with no such indication of a line of dehiscence.
2. Seta short and the foot deeply embedded in the gametophyte which closely envelops it.	2. Seta long and the foot not deeply embedded in the gametophyte.

Acknowledgments.

Summary.

A new species of mosses, *Physcomitrellopsis indica*, Dixon, is described in the present paper. This is the first record of the genus in India. It is a fairly big and gregarious moss with serrate leaves. It differs from the only other species *P. africana*, Broth. and Wager, chiefly in having an oblate spherical capsule with a distinct line of dehiscence which is entirely absent in the elliptic capsule of the African form.

The genus *Physcomitrellopsis* is distinguished from *Physcomitrella* by the fact that the calyptra is deeply lobed and covers the whole capsule in the young condition.

Literature.

1. BRÜHL, P. (1931)—A census of Indian Mosses. Records of the Botanical Survey of India, vol. XIII, Nos. 1 and 2.

2. CAMPBELL, D. H. (1928)—The structure and development of mosses and ferns.

3. DIXON, H. N. (1922)—Some new genera of mosses. Journ. of Botany, vol. LX, p. 107.

4. DIXON, H. N. (1924)—Student's handbook of British Mosses.

5. ————(1925)—Mosses from the Mount Everest Expedition 1924. The Journal of Botany.

6. ————(1926)—Mosses collected in Gilgit, etc. by J. Garrett and W. Lillie. Records of the Botanical Survey of India, vol. IX, No. 5.

7. ————(1930)—Addition to the moss flora of the N. W. Himalayas. Annales Bryologici, vol. III.

8. ————(1930)—Notes on Thwaites's Ceylon mosses. Journ. of Bot.

9. ————(1931)—New genera of Asiatic Mosses. Journ. of Bot.

10. ENGLER AND PRANTL (1924)—Natürliche Pflanzenfamilien. 10. Bd. New Ed.

Explanation of the Plates.

PLATE I.

Fig. 1. A single plant. Nat. size.

Fig. 2. Upper part of leaf.   ×70.

Fig. 3. Margin of leaf to show the serration caused by individual cells.   ×270.

Fig. 4. T. S. of stem with a branch and crowded leaves.   ×70.

Fig. 5. Line of dehiscence on capsule with lower cellwalls thickened.   ×420.

Fig. 6. Single stoma with annular guard cell.   ×420.

Fig. 7. Apophysis with a few stomata.   ×270.

PLATE II.

Fig. 8. T. S. fertile head.   ×270.

Fig. 9. Basal part of mature archegonium. Ventral canal nucleus not seen.   ×420.

Fig. 10. Young archegonium.   ×420.

Fig. 11. Entire antheridium.   ×420.

Fig. 12. Club-shaped paraphysis.   ×420.

Fig. 13. Capitate paraphysis.   ×420.

PLATE III.

Figs. 14–17. L. S. sporogonia at various stages of development.   ×70.

Fig. 18. Embryo with upper and lower apical cells; middle portion left blank.   ×420.

Fig. 19. Archesporium magnified.   ×420.

Fig. 20. Wall of the capsule, showing the point of dehiscence.
Enlarged from the section shown in fig. 15.   ×420.

Fig. 21. Basal part of the foot embedded in the gametophyte, show-
ing tip cells with dense contents.   ×270.

Figs. 22–24. Serial sections of an antheridium.   ×420.

PLATE IV.

Figs. 25–28. Outlines of the sporophyte at various stages of develop-
ment. ×90.

PLATE V.

Fig. 29. Composite figure to show the general disposition of the sex
organs and the sporophyte. Somewhat diagrammatic.

Fig. 30. Spores. Murications are diagrammatically drawn.   ×350.

Fig. 31. A dehisced capsule showing the regular arrangement of the
wall cells, especially in the operculum.   × ca. 80.

8

9.   10.   11.   12.   13.

FIG.25

FIG.26

FIG.27

FIG.28

Plate V.

GUPTA—*Physcomitrellopsis Indica.*

Fig. 29

Fig. 30
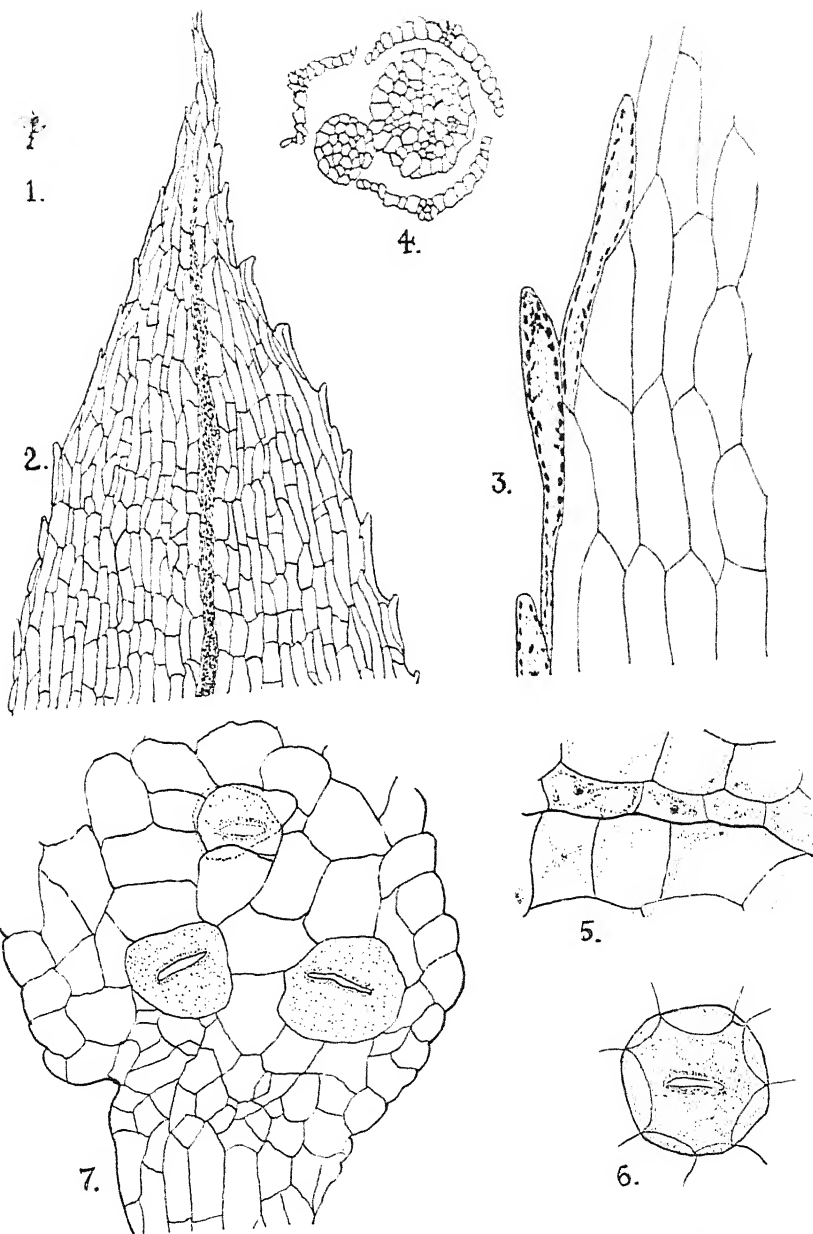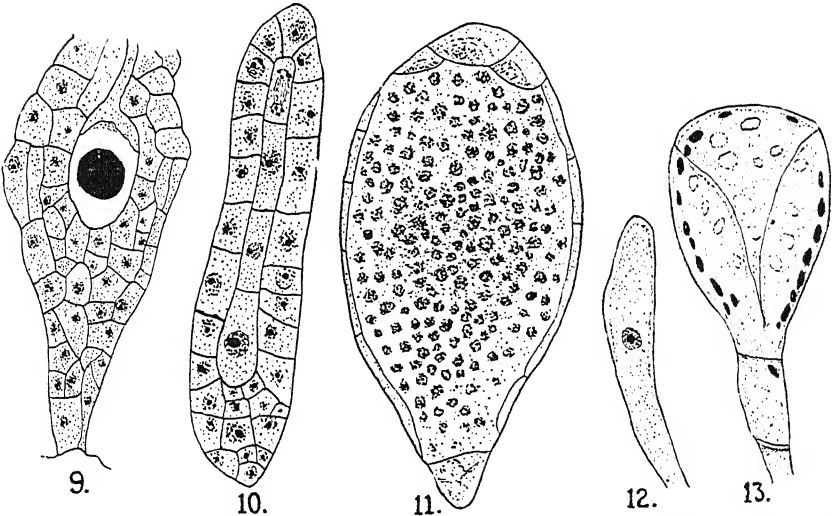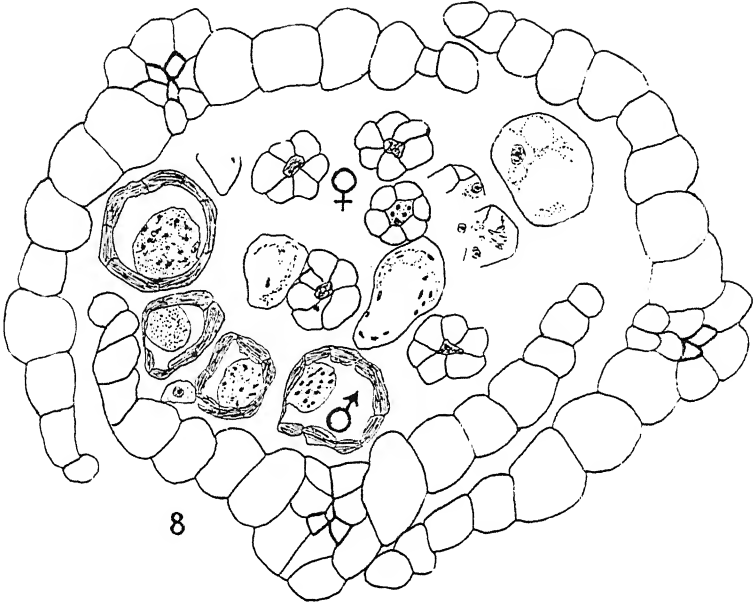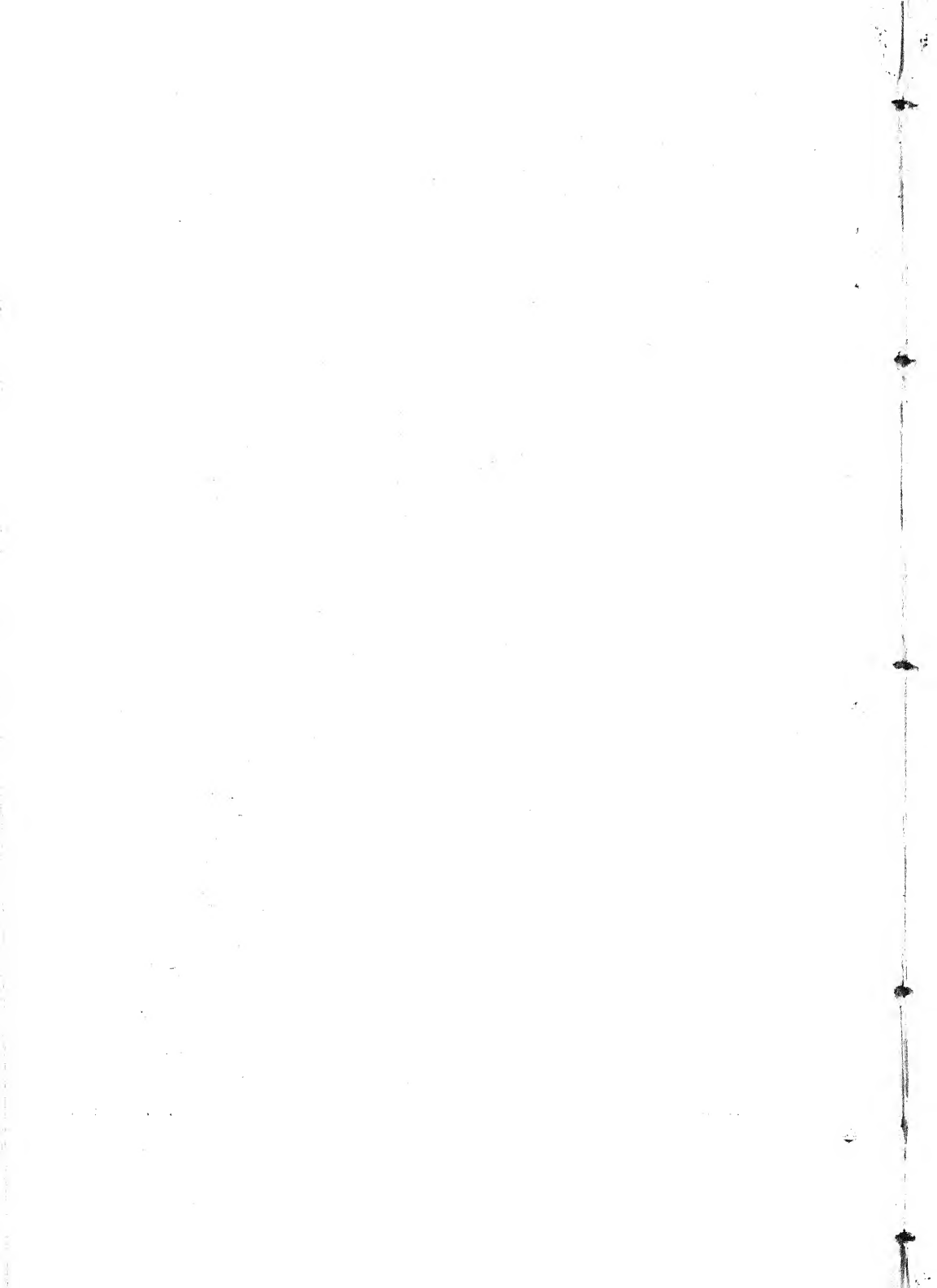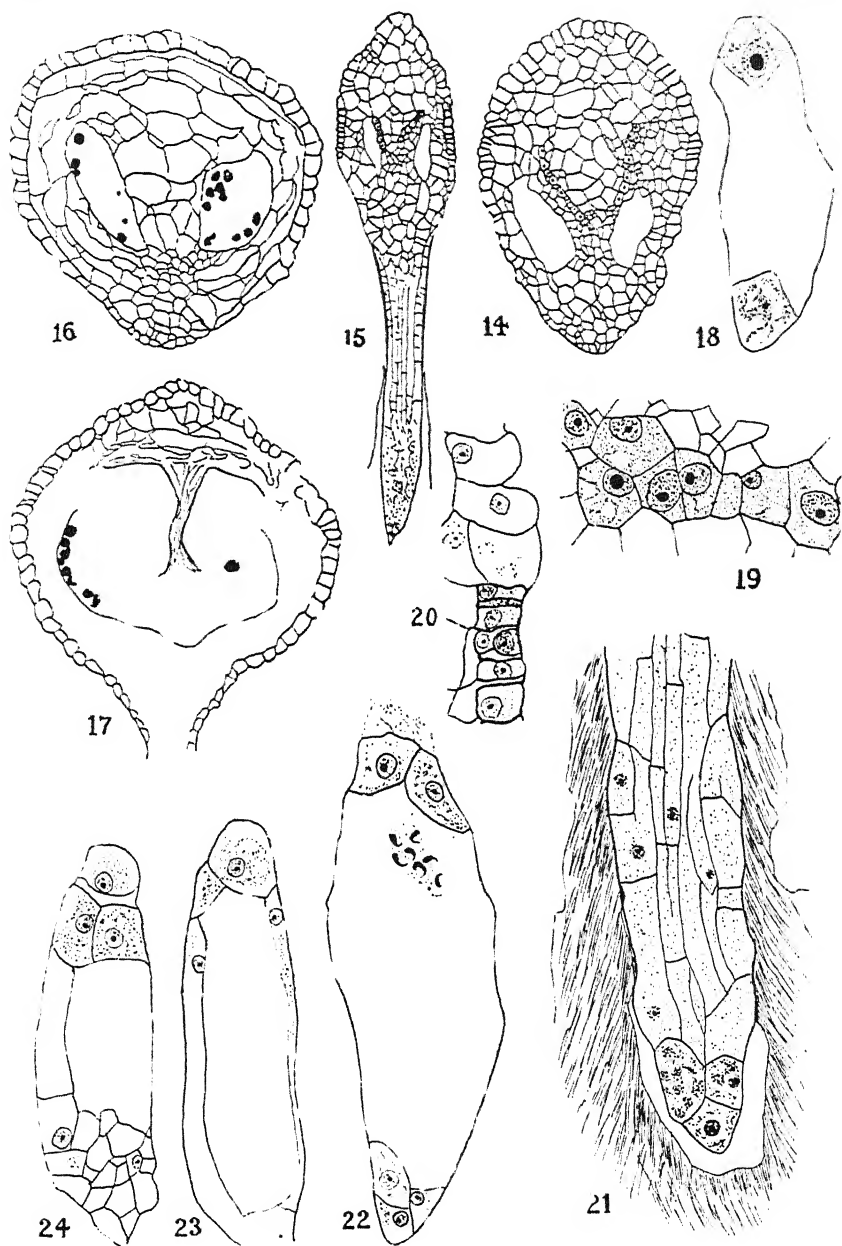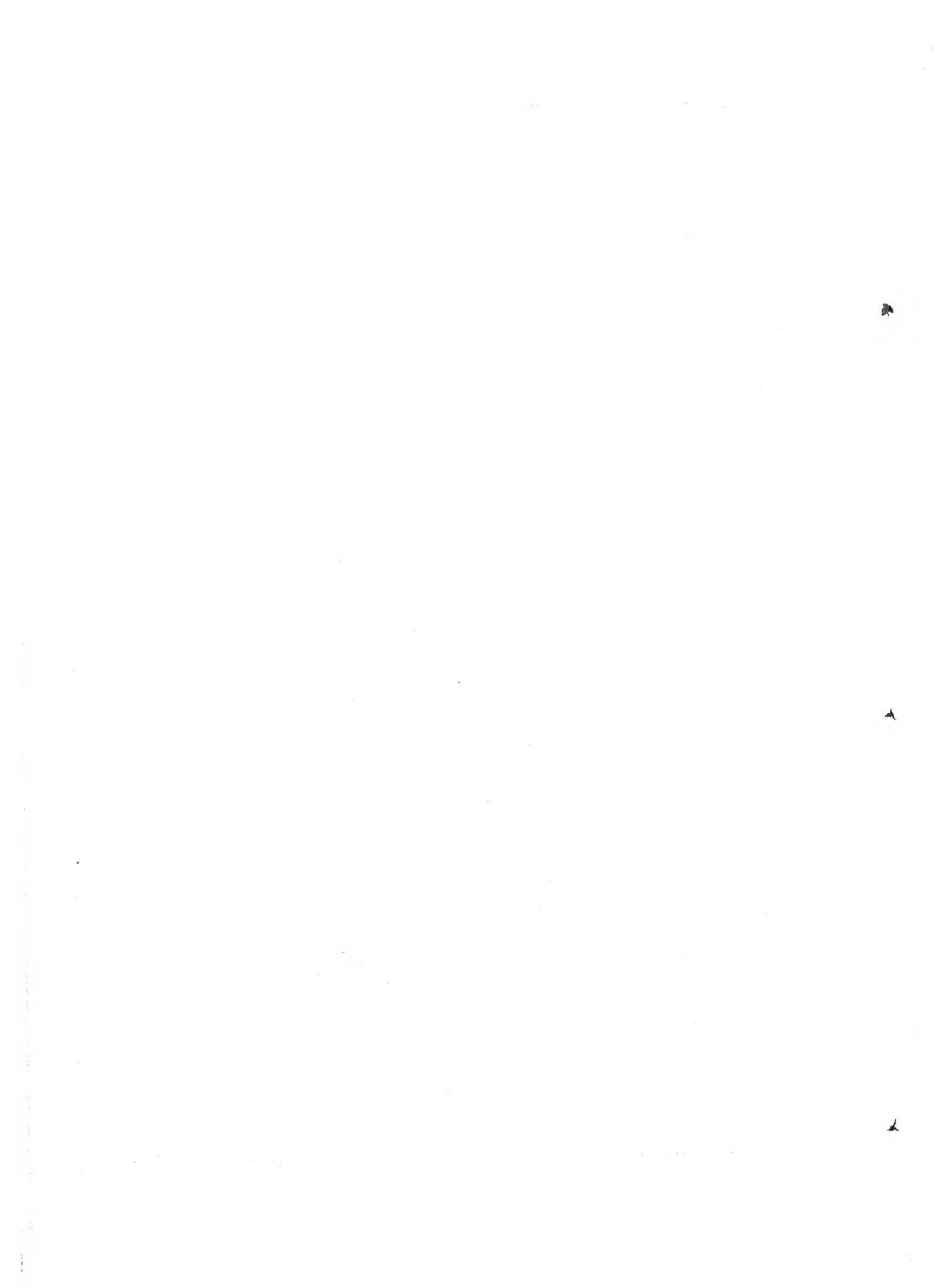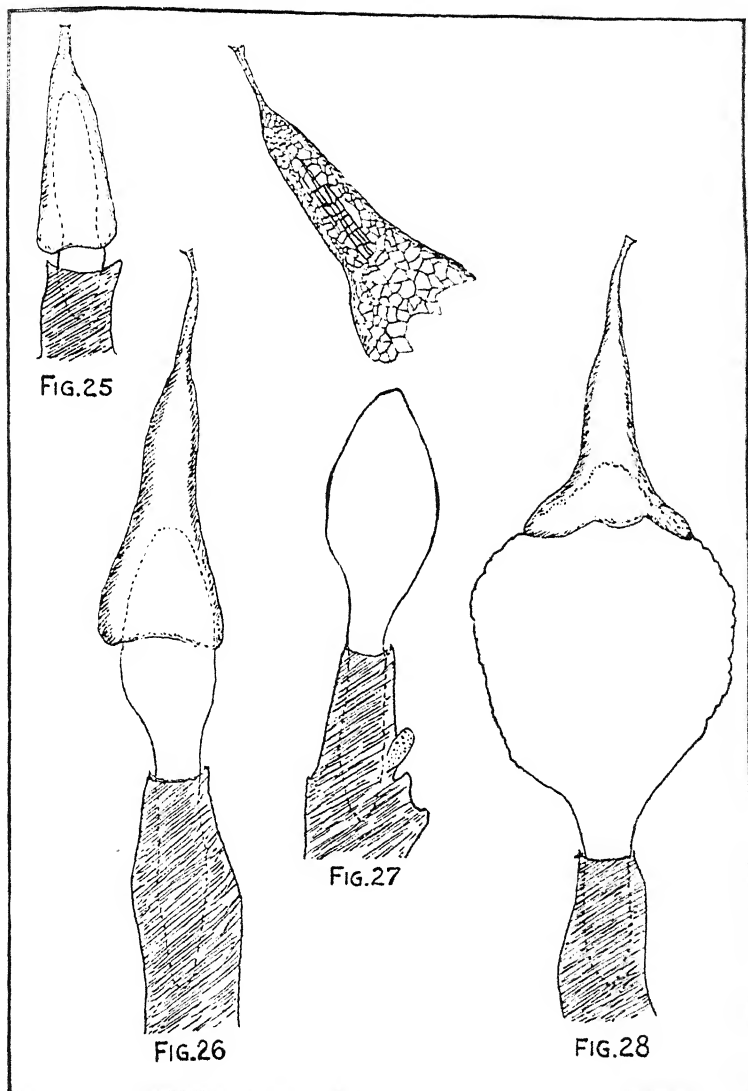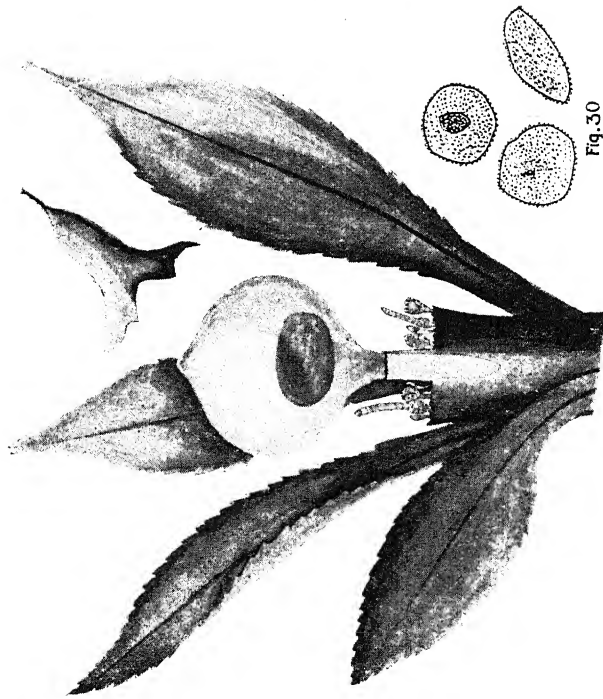
Fig. 31.

K. M. G. *phot.*; J. N. *del.*
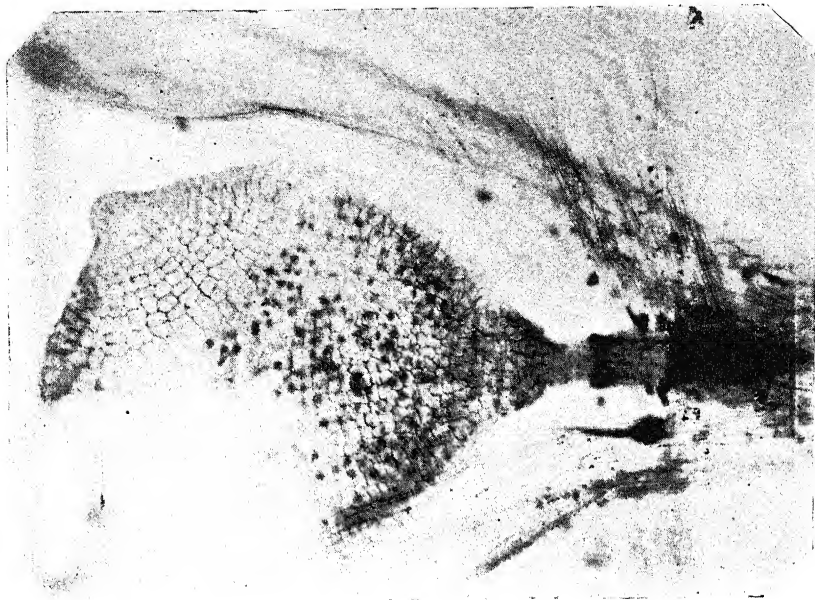
NOTES ON STAINING WITH IRON-ALUM HAEMA-TOXYLIN

BY

PANCHANAN MAHESHWARI, D.Sc.

In a recent paper TUAN[1] has given us a new method of Haematoxylin staining, much superior to the older methods. We have given this a trial for more than a year on various kinds of plant material from Algæ to Angiosperms and never failed to get good results. The essential point in the method is that a saturated solution of picric acid in water is used for the destaining process in place of Iron-Alum. The technique is so simple that any one can get good results after a little practice. A short schedule of the method, as we have been following it, is given here:—

1. After the sections have been brought down to water, mordant them by keeping the slides in a 2 per cent solution of Iron-Alum for about 15-30 minutes. In the months of May and June when the temperature rises considerably in North India, even 8-10 minutes are sufficient for most material. A longer immersion in the mordanting fluid is neither necessary nor desirable, as this leaves a disagreeable brownish colouration on the slide. But, for Algæ which are to be stained whole, about an hour will not be too much.

As is well known, only good Iron-Alum crystals of pure violet colour should be used for the solution, otherwise it leaves a precipitate on the slide which is impossible to wash off. There is, however, a lot of difficulty in keeping this reagent in tropical climates. In the hot months of summer, the crystals become brownish and melt in their own water of crystallisation. The only remedy is either to keep the bottle in a refrigerator, or prepare a sufficient quantity of the solution beforehand. This should, however, be filtered before use. Even the solution deteriorates in about 2 months from the time of making.

2. Wash the slides in running water for about 15 minutes.

3. Stain in $\frac{1}{2}$% Haematoxylin. Most authors use a previously prepared solution which has been allowed to ripen for at least 3 weeks. This does not seem to be necessary with some Haematoxylins at least. We have used Coleman and Bell's Haematoxylin without any ripening whatever and obtained excellent results.

---

[1] TUAN, HSU-CHUAN. Picric acid as a destaining agent for Iron-Alum Haematoxylin. Stain Technology: 5: 185-188. 1930.

Perhaps the best plan is to weigh the Haematoxylin and dissolve it in the requisite quantity of hot tap water. The solution gives a rich red colour. If it is allowed to stay for a day or two in an uncorked bottle, the colour darkens. This should now be filtered and it is ready for use.

Some investigators dissolve a large quantity of the stain in alcohol and from this stock solution they make small quantities as required from time to time. This is not necessary, as the crystals dissolve in less than an hour in hot water and the stain can be used soon after.

No exact time limit can be given regarding the immersion of slides in the Haematoxylin solution. For most things half an hour is enough. Sections of *Limnophyton obtusifolium* flowers stained sufficiently in less than 5 minutes. *Azolla* seems to take a longer time, about an hour or so. For Algæ to be stained in bulk, about 2-4 hours' immersion is necessary.

4. Wash in water for about 5 minutes.

5. Destain in a saturated solution of picric acid. It is not necessary to weigh; put enough picric acid in a bottle and fill it with water. Shake it vigorously for some time and then allow the undissolved acid to settle at the bottom. The solution can be decanted from time to time.

It will be necessary to take out a slide from time to time from the staining dish, rinse it in water and examine it under the microscope. If the destaining has been satisfactory, the chromosomes should remain black and the cytoplasm should not have more than a bluish tinge. If one is staining large quantities of the same material, he will soon get an approximate idea of the time required for destaining.

6. Wash the slides in running water for about 30 minutes to get rid of the picric acid. Dehydrate and mount as usual.

The advantages of this method are:

1. The whole process can be completed in about 2 hours for one batch of slides.

2. The slides show no brownish colouration, seen very often if the sections are destained in Iron-Alum.

3. Being milder in its destaining properties, picric acid has a more precise action which can also be easily controlled.

BOTANY DEPARTMENT,
AGRA COLLEGE,
AGRA, INDIA,
25th January, 1933.

EMBRYOLOGICAL AND CYTOLOGICAL STUDIES IN *NOLANA ATRIPLICIFOLIA* AND *NOLANA PROSTRATA* *

BY

SOROJINI DATTA, M.A. (CAL.), M.Sc. (MAN.);

Department of Botany, Bethune College, Calcutta.

(With 4 Plates).

Contents.

Introduction.

There appears to have been some difficulty in determining the systematic position of the order to which the plants belong.

The family is variously classed by different authorities:—

In 1852 M. Dunal (5) divided the family Solanaceæ into two tribes: the Nolanaceæ and the Solanaceæ. In 1857 John Miers (11) criticised the above division of M Dunal and regretted that he should have associated the Nolanaceæ with the Solanaceæ, from which they differ essentially in the structure of the pistil and the fruit. On page 44 of the same work he summarised the views of the previous authors and of himself thus:—"Schlechtendal in 1832 (Linnæa VII. 12) pointed out the analogy that exists in the Genus Nolana to the families of the Boraginaceæ and Solanaceæ, admitting its greater affinity with the former, ... Dr. Lindley, who first proposed this order in 1833, placed it near the Convolvulaceæ. G. Don (1837) was, I believe, the first who decidedly associated the

Nolanaceæ as a tribe of the Solanaceæ (Dict. N. 399), but he offered no reasons for this union. Endlicher in his 'Genera Plantorum' followed the views of Dr. Lindley, in attaching this group as a suborder of the Convolvulaceæ. Brongniart (1843) adopted the same views in regard to the affinity of the Nolanaceæ. A. de Jussieu (1844, Cours Elémentaire) equally confirmed the ideas of the before mentioned botanists, in placing the Nolanaceæ in contiguity with the Dichondreæ, between the Boraginaceæ and Convolvulaceæ. In 1845 I adduced many facts and several additional reasons, why the Nolanaceæ should be placed in the system following the Boraginaceæ (Lo. Jo. Bot. IV. 366 et huj. op i, 46), which position was confirmed in the following year under the arrangement given by Prof. Lindley (Veg. Kingd. 654), where this order is placed in his Echial alliance with the Boraginaceæ, Labiateæ and others."

In 1876 Bentham and Hooker (1) placed the group Nolanaceæ as a separate tribe under Order CXIII Convolvulaceæ of the Cohort Polemoniales, and the series Bicarpellatæ. Engler and Prantl (7) in 1895 described Nolanaceæ as a separate group and placed it just before the Solanaceæ. The family included three genera, viz :—(1) Nolana L, (2) Alona Lindl, and (3) Dolia Lindl. The genus Nolana L was again sub-divided into two sections, viz :—

(1) Eunolana Miers............ e.g. *Nolana Prostrata ;*

(2) Sorema Lindl............... e.g. *Nolana Atriplicifolia.*

The systematic position is further described as follows :—

"Verwandtschaft. Die Familie der N nimmt systematisch eine sehr interessante Zwischenstellung zwischen den Convolvulaceæ, Boraginaceæ und Solanaceæ ein. Mit den Convolvulaceæ, mit denen sie von Bentham u Hooker vereinigt wurde, verbindet sie eine Ahnlichkeit im Baue de Blkr. und der 3–5 blattrige Frkn, welcher eine Anlehnung an den Mancher Convolvulaceæ zeigt. Die Ähnlichkeit mit den Boraginaceæ beruht in der eigentümlichen Fruchtbildung. Am grössten ist zweifellos die Verwandschaft mit den Solanaceæ, denen die Familie auch von Dunal und neuerdings von Baillon zugezogen wurde. Die Übereinstimmung liegt in der Knospenlage der Blkr, in der verzweigung des Stengels, in der Blattstellung und in Baue des. S. Der Vollkommen abweichende Fruchtbau lässt aber eine vereinigung der beiden Familien Unmöglich zu. Den Anschluss vermitteln die N. Gattungen Alona und Dolia und die Solanaceæ—Gattungen Phrodus und Grabowskia."

In 1895 Warming (19) placed Nolanaceæ as Order 2 under family 30–Personatæ of Section 2–Tetracycliceæ. He further

observed that "These (Nolanaceæ) most resemble Convolvulaceæ in the corolla, but the Solanaceæ in their branching and leaf-arrangement (in pairs, etc.) ". In 1911, Wettstein (20) classified Nolanaceæ as the 4th family of the 6th Series-Tubifloreæ of the second Subclass-Sympetaleæ. In 1925 Rendle (12) placed Nolanaceæ as a separate family VI, under the order 4—Tubifloræ. He says, "Bentham and Hooker included it in Convolvulaceæ, an affinity with which is suggested by the form and aestivation of the corolla, and the pentamerous ovary. The separation of the fruit into mericarps recalls Boraginaceæ. It is, however, probably most nearly allied to Solanaceæ, with which it was united by Dunal and Baillon, but from which it is distinguished by the structure of the fruit." But in 1926 Hutchinson (8) in his proposed phylogenetic system included this group in the family 251—Convolvulaceæ, under the order 74,—Solanales.

From the above summary it will be seen that the systematic position of the Nolanaceæ as given by different authorities has possibilities of affinities with the following three groups, viz :—(1) Solanaceæ; (2) Boraginaceæ and (3) Convolvulaceæ.

The cytological and embryological investigations of *Nolana Atriplicifolia* and *Nolana Prostrata*, belonging to the family Nolanaceæ, have been undertaken with a view to elucidation of the systematic position of the family. Some reciprocal hybrids of these two species have also been worked out before their fertilisation to find out whether or not there is any abnormality in their chromosomes.

Materials and Methods.

The different stages of developing fruits of *Nolana Atriplicifolia* and *Nolana Prostrata* were fixed in the months of August and September, during the flowering season, in the Cambridge Botanic Garden and the Kew Gardens.

After comparative tests of some fixatives the best result has been obtained by dipping into Carnoy's fluid for about 3 to 5 minutes and then in the Chrom-Acetic-Formalin solution of full strength (10) with or without an addition of a little solid maltose. Root tips for chromosome study have been fixed in Chrom-Acetic-Formalin solution of full strength.

Sections parallel to the flat surface of ovules taken out from the carpels were cut 8 microns in thickness. Root-tips were cut 6 microns in thickness.

For staining, the following combinations have been tried :—

  (1) Iron Hæmatoxylin and Light Green.
  (2) Safranin and Light Green.
  (3) Safranin and Delafield's Hæmatoxylin.
  (4) Gentian Violet and Light Green.
  (5) Cyanin and Erythrosin.

Iron Hæmatoxylin and Light Green combination gave the best result. Gentian Violet stained the chromosomes well in the background of green stained cytoplasm with Light Green.

Living embryos of *Convolvulus Tricolor* and entire embryos from fixed material of Nolana were dissected out as a whole under a binocular dissecting Microscope. For permanent preparations a few of these dissected embryos, specially the advanced stages, were stained with Eosin in 95 per cent alcohol and after dehydration were permanently mounted in Canada Balsam. But most of these living embryos dissected out as a whole, were placed first in dilute glycerine for 15 to 20 minutes and then mounted permanently in glycerine jelly coloured with Erythrosin. Some embryos of this species were fixed like those of the Nolana species and sections were cut 8 microns in thickness.

The illustrations of embryos of *Nolana Atriplicifolia* and *Nolana Prostrata* are drawn under $\frac{1}{12}$ Homog. Imm. objective apert. 1.30, eye-piece IV. Magnification is 960 times. Chromosomes are drawn under the same objective with eye-piece 18 at a magnification of 2600. Some embryos of *Convolvulus Tricolor* are drawn under ordinary high power objective 7A with eye-piece IV giving a magnification of 520. Some figures of dissected out embryos of this Convolvulus species and *Nolana Atriplicifolia* are drawn under low power objective 3, eye-piece IV and with a magnification of 97. All figures are drawn under Camera Lucida on the drawing table on same level with the Microscope stage.

Embryological Data.

In both the species *Nolana Atriplicifolia* and *Nolana Prostrata* the unfertilised embryo sac is a normal one.

(I) THE FILAMENTOUS PRO-EMBRYO.

In both the species the first division of the egg after fertilisation is by a transverse wall giving rise to an apical cell CA towards the embryo sac and the basal cell CB towards the micropylar end (Fig. NP1 & Fig. NA 2). The apical cell CA is well differentiated from the very beginning from the basal one, it being shorter and somewhat rounded in shape and having richer protoplasm. This cell appears to

take an essential part in the formation of the embryo. The basal one CB is elongated and flattened at the base and forms the suspensor. Many endospermic nuclei by now are formed in the embryo sac.

The two cells CA and CB of this bicellular proembryo then segment transversely to give 4 superposed elements viz: l and l' from CA and m & ci from CB (Figs. NP 3 & NA 4). The fig. NA 2 shows that the apical cell CA divides first.

A six-celled stage of the filamentous pro-embryo has been observed in *Nolana Prostrata* (Fig. NP 5); 5 cells contain richer protoplasm and the basal one is rather large and haustorial. The clear evidence of the origin of two cells from the l' (the penultimate of the tetrad) is well-marked in the figure NA 4 where the cell is in mitosis and probably two terminal cells (Fig. NP 5) have been derived from the element l.

The details of further segmentation of the 6-celled pro-embryo is probably variable (Figs. NA 6, NP 7, NA 8, NP 9), for the maximum number of cells in the filamentous pro-embryo seems not quite uniform (cf. Figs. NA 6, NP 7, NA 8, NP 9). But these differences, however, do not appear to affect the subsequent segmentation of the embryo proper.

II. SEGMENTATION OF EMBRYO PROPER.

The three apical cells, which may conveniently now be named I, II, III, take part in the formation of the embryo proper. The penultimate cell II has tendency to divide first (NA 8), then the terminal one, No I, starts to divide. The III cell is always later in division than these two.

The ultimate fate of each tier is as follows :—

    I the terminal one gives origin to cotyledon and stem apex.

    II the penultimate one gives origin to hypocotyl and radicle.

    III the third tier gives origin to middle of the root cap.

In the segmentation of the embryo proper the first two terminal layers (Figs. NA 6, NP 7, NP 9) enlarge and divide vertically to give rise to a quadrant stage of the embryo. The fig. NA 8 shows the penultimate one II starting to divide vertically. Then these 4 cells of the embryo in these two tiers I and II segment by walls at right angles to the first walls to cause an octant stage of the embryo (Fig. NP 11). In the process of the formation of this octo-cellular stage of the embryo the fig. NP 10 shows that the two cells in the penultimate Cell II first have divided to cause 4 cells.

III. EARLY DIFFERENTIATION OF TISSUES.

    (a) *Formation of dermatogen.*—The figs. NA 12, NP 13, NP 14 show that after the octant stage of the embryo (i.e. 4 cells in the tier I

and 4 cells in the tier *II*) the dermatogen begins to be differentiated in the two terminal tiers *I* and *II* by periclinal segmentation. The differentiation in both the tiers is nearly simultaneous, so that the dermatogen is soon completed except that of the root tip, *i.e.* in the third tier *III*.

(b) *The segmentation of hypocotyl from radicle.*—After the differentiation of the dermatogen the interior cells of the penultimate layer *II* (Figs. NA 17, NA 18, NA 19, NA 20b) divide transversely to separate the cells of hypocotyl from those of the radicle, forming two layers r and h one above the other. The outer cells, i.e. the dermatogen-forming ones and in the penultimate region—the tier *II*, divide longitudinally. Figs. NA 20a and NA 20b show two adjoining sections through the same embryo and 5 of the 8 dermatogen cells can be seen. Transverse divisions in this outer-ring then separate two layers. corresponding to the two tiers h and r already formed in the central cells, i.e. representing the dermatogen of the hypocotyl and radicle respectively (cf. Figs. NA 21, NA 23, NP 22, NA 24, NA 26).

(c) *Early segmentation in the cotyledon tier I* :—After the formation of two layers for separation of the hypocotyl from the radicle in the tier *II*, the 4 inner elements in the tier *I* cause two layers one above the other by establishing transverse divisions (Figs. NA 19, NA 21, NP 22, NA 23, NA 25, NA 26). The outer cells i.e. the dermatogen, also divide anticlinally to give rise to two layers (Figs. NA 21, NP 22, NA 23, NA 24, NA 25, NA 26).

(d) *Further development in the hypocotyl and radicle regions.*— In the progress of further growth in both the hypocotyl and radicle regions the inner cells in most cases divide longitudinally and then establish transverse divisions (Fig. NA 23, NA 24, NA 26, NP 27, NA 29). The Figs. NA 23 and NA 24 show that division in the radicle-forming layer r tends to precede those in the hypocotyl region. The dermatogen cells of h and r divide anticlinally to keep pace (Figs. NA 29, NA 30, NP 31). The inner cells in the radicle-forming layer r sometimes have oblique divisions (cf. the left side inner cell in r, i.e., in the lower half of the tier *II* in Fig. NA 25). In this case there are thus formed two elements very dissimilar in shape and size, the large outer one separating the smaller inner one both from the dermatogen and the layer *III*. The inner smaller cell engenders the cells entering into the formation of the central cylinder and the outer portion gives origin to the cortex of the root. When the original inner cells have longitudinal and transverse (instead of oblique) divisions, the central cylinder is differentiated from the cortex of the root only after the formation of a fairly massive tissue (Figs. NA 30, NP 31, NA 32).

(e) *The formation of root cap from the tier III.*—Up to the formation of the octant stage of the embryo (i.e. the 4 cells in the tier *I*, and 4 cells in the tier *II*) this layer *III* only comprises a single cell (cf. Figs. NP 11, NA 12). In some cases (Figs. NA 17, NA 21, NA 23) this layer does not divide for a long time. But often along with the differentiation of the dermatogen (Fig. NP 14, NA 15, NP 16) it gives rise to a tier of 4 cells by two walls at right angles to one another. Later on in the advanced stage the details of segmentation in these 4 elements vary. In many cases they divide obliquely (Figs. NA 29, NA 30, NP 31) but longitudinal and transverse segment-ations have also been observed (Figs. NP 27, NA 28, NA 39, NP 40). In each case the subsequent mode of segmentation is different though the final results appear to be similar. In the case of horizontal division the upper layers towards the embryo initiates at once part of the calyptrogen and the layer towards the suspensor forms an external layer of the middle part of the root cap. When the division is longitudinal or oblique these cells are believed to divide again trans-versely to give rise to two layers of cells. Their formation in details have not been observed, but it seems in each case that the ultimate result is the same to cause two layers of cells one above the other segmenting off calyptrogen initials from the external part of the cap. To the central portion of the rootcap lateral parts are probably added, being originated by tangential divisions of the lowest epidermal cells of the basal region, viz : r of the tier *II* (Fig. NA 33).

(f) *Suspensor.*—The suspensor ultimately consists of a vertical row of 4–9 cells (Figs. NP 7, NA 8, NP 10, NA 12, NP 13–14, NA 15, NP 16, NA 17, NA 19, NA 21, NP 22, NA 23, NA 26, NP 27, NA 30, NP 31, NP 34). Occasionally the cell nearest the root cap may divide vertically to form a tier of two (Figs. NA 26, NA 29, NA 30, NA 32, NP 34). Sometimes an oblique division occurs in the basal haus-torial cell (Figs. NA 8, NA 21, NA 26, NA 30).

(g) *Formation of Cotyledon & Stem-apex.*—In the further development of the cotyledonary tier *I* the inner cells of the two layers divide longitudinally and transversely to add new cells in the formation of cotyledonary protuberances (Fig. NA 29, NA 30, NP 31). The dermatogen also divides to grow along with the development of the inner parts of the cotyledonary tier. In a well-advanced stage these two cotyledons bulge out (Fig. NA 35) but become curved in the ripe seed (NA 36).

The stem-apex is presumably also formed from the apical tier *I* at the base of the groove between the cotyledons (cf. Figs. NA 35 and 36). But in dissected out old embryos, (Fig. NA 35) and even in the

sections of the ripe seed (NA 36) the stem-apex is not seen to be originated. So it seems it does not develop until after germination.

IV. ILLUSTRATION OF ABNORMAL CASES.

There are some illustrations of abnormal embryo formation both in *Nolana Prostrata* and *Nolana Atriplicifolia*. The Fig. NP 37 shows a long filamentous structure of 10 cells in 9 tiers the apical one having divided abnormally. The Fig. NA 38 shows the division of the 4th cell of the pro-embryo in addition to the third. In the Figs. NA 39, NP 40, NA 41, the first tier has remained inactive whereas the second and third tier are quicker in speed of segmentation, and cell division seems to have been without order.

Discussion.

Such are the general data which emerge from the study of the embryonic development in the species *Nolana Atriplicifolia* and *Nolana Prostrata* belonging to the group Nolanaceæ. Brief comparison with available data for the other families mentioned in the introduction may now be made.

COMPARISON OF NOLANACEÆ WITH SOLANACEÆ.

In his "Embryologie der Angiospermen" Dr. Karl Schnarf (14) has described that in the Solanaceous type of embryology the apical cell CA of the bicellular pro-embryo takes an essential part in the formation of the embryo and the cells derived from the basal one CB either take a very little part or no part at all, being only suspensor-producing cells (p. 397). In these two species *Nolana Atriplicifolia* and *Nolana Prostrata* we have already seen that the same law is followed in the embryo formation. In them the basal cell CB does not take part at all in the segmentation of the embryo proper.

Souèges (16) records the following facts about the embryo formation in Solanaceæ :—

(1) In the Solanaceæ in general, from the two cells CA and CB of the bicellular pro-embryo develop 4 superposed cells (l, l', m and ci). Only in Datura, the apical cell CA occasionally divides vertically and the basal cell CB segments horizontally causing 4 cells arranged in three layers. In both the Nolana species the 2-celled and the 4-celled filamentous pro-embryo have been found to have originated in the same way as in all the species of Solanaceæ other than Datura.

(2) In the types that may be considered as normal in the Solanaceæ the apical cell CA segments before the basal one. The evidence of the apical cell CA in mitosis in *Nolana Atriplicifolia* (Fig. NA 2) clearly shows the same thing.

(3) The length of the filamentous pro-embryo is different in different species of Solanaceæ. In the majority of cases, viz:—in *Nicotiana Sanguinea* Link., *Nicotiana Tabacum* L., *Hyoscyamus Niger* L., *Datura Stramonium* L., *Atropa Belladona* L., *Solanum Nigrum* L., the filamentous pro-embryo is a short one of 4 cells only. There are, however, cases of 6-to 8-celled filamentous pro-embryos in a few species. In *Solanum Dulcamara* L. a 6-celled pro-embryo occasionally develops though the usual number of cells is 4. In *Solanum Villosum* Moench. a 6-celled filamentous pro-embryo is the rule, while in *Solanum Sisymbrifolium* 6, 7 and 8-celled ones occur. The origin of the 6-celled pro-embryo in *Solanum Dulcamara* and *Solanum Sisymbrifolium* differs. In *Solanum Dulcamara* 4 apical cells of the 6-celled pro-embryo are derived from the apical cell CA and 2 basal cells from the basal one CB of the bicellular pro-embryo, whereas in *Solanum Sisymbrifolium* 2 apical cells are contributed by the apical cell CA and 4 basal cells by the basal one CB. The occasional production of 7 and 8-layered pro-embryos in *Solanum Sisymbrifolium* is due to the variable direction of division in the two apical cells (1 and 1') of the 6-celled stage. This may be longitudinal, oblique or transverse; when both 1 and 1' divide transversely an 8-tiered filament results. These differences, however, do not appear to affect the subsequent segmentation of the embryo proper.

In both the Nolana species the pro-embryo is a six-celled one and probably developed like *Solanum Dulcamara*. As in *Solanum Sisymbrifolium* so in Nolana the details of further segmentation of this 6-celled filamentous pro-embryo seems to be variable but the differences do not affect the subsequent division of the embryo proper.

(4) In the Solanaceous type the three end cells (called here I, II, III) take part in the segmentation of the embryo proper in the following way :—

I gives rise to the cotyledon and stem-apex.

II   „   „ „ „   hypocotyl and radicle.

III   „   „ „ „   middle part of the root cap.

In Nolana the three end cells take part in the formation of the embryo in the same manner.

(5) In the Solanaceæ, viz., In *Nicotiana* species, *Hyoscyamus Niger*, *Solanum Nigrum*, *Solanum Villosum* Moench and *Solanum Sisymbrifolium* two cells in each of the three tiers I, II, III give rise to 4 cells by walls at right angles to one another. In all, the penultimate tier II first starts to divide and segments more rapidly than the others, then the end one and lastly the third III. The same order of segmentation has also been observed in *Nolana Atriplicifolia* and *Nolana Prostrata*.

(6) In *Nicotiana Acuminata, Hyoscyamus Niger, Solanum Nigrum, Solanum Dulcamara* in the octant stage of the embryo (*i.e.* 4 cells in the tier I and 4 cells in the tier II) the dermatogen begins to be differentiated by periclinal divisions.   On the other hand, in *Solanum Nigrum* some cells in the tier I sometimes segment transversely before the differentiation of the dermatogen, while in *Atropa Belladona* cells in the tier II occasionally divide transversely to separate the hypocotyl layer early from the radicle-forming one before the separation of dermatogen.

In both the Nolaña species resemblance is to the average type and not to *Solanum Nigrum* or *Atropa*, the dermatogen being formed in both the tiers I and II directly after the octant stage of the embryo.

(7) In *Nicotiana Acuminata, Hyoscyamus Niger* and *Solanum Nigrum* after the differentiation of the dermatogen from the inner cells in the tier II, the next wall is transverse in these cells to cause two layers one above the other for the separation of the hypocotyl from the radicle.   On the other hand, in Hyoscyamus the separation of the hypocotyl from the radicle is sometimes delayed whilst in Atropa it may occur prematurely (cf.   para 6, above.)   In both *Nolana Prostrata* and *Nolana Atriplicifolia* the hypocotyl and radicle regions are laid down as in *Nicotiana Acuminata* and *Solanum Nigrum*.

(8) In the segmentation of the cotyledonary tier after the formation of dermatogen in *Solanum Nigrum* and *Atropa Belladona*, transverse walls are established in the 4 inner elements and anticlinal divisions in the dermatogen to cause two layers one above the other. On the other hand, in Nicotiana and Hyoscyamus the subepidermal tissue of tier I remains as a single layer until a late stage, longitudinal walls only being formed.   Tangential divisions occur in *Solanum Villosum* and occasionally elsewhere.

In both *Nolana Prostrata* and *Nolana Atriplicifolia* neither the tangential nor the vertical segmentation have been observed.   In all cases as in *Atropa Belladona* there are transverse divisions in the inner cells and anticlinal divisions in the dermatogen producing two superposed layers directly.

(9) There appears to be nothing remarkable in the further growth of the cotyledon and hypocotyl regions in any of the Solanaceous species, but details are given of the further development of the radicle portion.   There are differences in the origin of the central cylinder and cortex of the root.   In Hyoscyamus an oblique division in each of the 4 inner cells of r occurs to separate early the central cylinder from the cortex of the root.   In *Solanum Nigrum* and *Atropa Belladona* the same differentiation appears occasionally, but

more often the central cylinder is differentiated out only after a fairly massive tissue has formed. In Nolana species separation of the central cylinder sometimes appears early by periclinal division in the inner cells (fig. NA 25), but usually is late. There is therefore some resemblance to *Solanum Nigrum* and *Atropa Belladona*.

(10) In Solanaceæ sometimes the third tier divides slowly and does not do much for a long time. In Hyoscyamus and *Solanum Nigrum* 4 cells are formed in the tier III rather early even before the formation of the dermatogen in the other two. The process of segmentation in these 4 cells is variable. In Hyoscyamus they next divide horizontally, whereas in Nicotiana and *Atropa Belladona* they divide either obliquely or horizontally. However, in each case two layers are finally formed one above the other separating the elements of calyptrogen from the external layer of the root cap. The lateral portions are added to right and left of the central part of the root cap by tangential division of the epidermal cells of the base of the lower part of the tier III. In Nolana species 4 cells in the tier III sometimes have the same horizontal division (fig NA 28) as in some Solanaceous species. On the other hand in Nolana these cells occasionally divide vertically or obliquely. However, the general mode of segmentation in the development of calyptrogen and rootcap seems usually to be as described in Solanaceous species.

(11) In Solanaceæ the suspensor shows some diversity. In Nicotiana it consists of 3 to 5 cells. In Hyoscyamus and *Solanum Nigrum* it has five to six cells. In Datura its shape is clearly bulky. But in *Solanum Sisymbrifolium* the lower cells give rise to a suspensor which attains an unusual length of having 8 superposed cells. In Hyoscyamus and *Solanum Nigrum* one vertical division takes place in the cell near the root cap. In Nolana species the usual length of the suspensor is long, 4 to 9 cells. This is, however, not widely different from the condition in *Solanum Sisymbrifolium*. When the suspensor consists of 9 cells, the cells tend to be rather short and broad (figs. NA 21, NP 22). In other cases cells are rather long (Fgs. NP 16, NP 27). Like Hyoscyamus the cell nearest to the root cap occasionally divides vertically. The basal haustorial cell is always bigger in size than the others and sometimes divides obliquely.

The above comparison shows that from the very beginning of the growth of the fertilised egg to the developed embryo in *Nolana Prostrata* and *Nolana Atriplicifolia* there is a well-marked analogy with the embryo formation of the Solanaceous type. The chief difference is in the length of the suspensor but the points of likeness are more numerous and are of a more fundamental nature.

COMPARISON WITH BORAGINACEÆ.

Karl Schnarf (14) has placed *Myosotis Hispida* (Boraginaceæ) under his fourth group, the " Chenopodiaceen-Typus ". He has defined this type thus—"ca wird durch eine Querwand in zwei übereinanderliegende Zellen geteilt; an der Bildung des Keimlings hat ausser ca auch cb wesentlichen Anteil." So in Boraginaceæ the products contributed from ca and cb both take part in the formation of the embryo whereas in *Nolana Prostrata* and *Nolana Atriplicifolia* the product derived from ca only takes an essential part in the embryo formation.

Souéges (17) has recorded the following facts about Boraginaceæ with reference to *Myosotis Hispida* :—

(1) The two elements of the bicellular pro-embryo segment transversely to give 4 superposed cells l and l', m and ci. In *Nolana Prostrata* and *Nolana Atriplicifolia* and in the Solanaceæ the construction is the same.

(2) The 6-celled pro-embryo in *Myosotis Hispida* is formed by vertical division of the cell l' i.e., the penultimate one, but by oblique segmentation in the terminal one l. The latter thus gives two dissimilar elements a and b in the tier I.

Early enlargement and segmentation of the cells for the embryo proper and the oblique division in the tier l are peculiarities of *Myosotis Hispida*. So the ways of segmentation in the 4 cells l, l', m and ci of the pro-embryo in *Myosotis Hispida* clearly show that just after the 4-celled stage the analogy of the Nolanaceæ with the Boraginaceæ stops. (see also para 5 below).

(3) In the segmentation of the embryo proper in *Myosotis Hispida* the 4 elements l, l', m and ci take part in the formation of the embryo proper as follows :—

l engenders stem and cotyledon ;
l'   ,,   half i.e. the upper part, of hypocotyl :
m   ,,   half i.e. the lower part, of   ,,
ci   ,,   radicle + root cap + suspensor.

But in *Nolana Prostrata* and *Nolana Atriplicifolia* only three end cells take part in the formation of the embryo, and all these three (tiers I, II, III) seem to have originated from l, and l' contributed from the apical cell ca of the bicellular pro-embryo (cf. page 137).

(4) In Boraginaceæ in the four embryo-forming tiers subsequent divisions first appear in the terminal cell then proceed to the second, third and lastly to the fourth one. But in *Nolana Prosatra* and *Nolana Atriplicifolia* the penultimate (II) has a tendency to

divide first, then the terminal one (I) and lastly the third tier divides. So the order of segmentation in the embryo-forming cells in Nolana also differs from that in the Boraginaceæ.

(5) In Boraginaceæ the two dissimilar elements called a and b in the tier I (or l, see p. 144, para 2 above) segment by 2 walls to give rise to 4 elements occupying a tetrahedral structure in the tier I. The element which is thus placed at the top of the tier I has received the name epiphysis and engenders early the initials of the growing point of the stem and its epidermal layer. In *Nolana Prostrata* and *Nolana Atriplicifolia* the 2 cells in the tier I segment by 2 walls at right angles to one another to give rise to 4 elements all in the same plane. There is no sign of an epiphysis and therefore the mode of segmentation in the cells for the formation of stem tip and cotyledons in the tier I in Nolana does not agree at all with that in *Myosotis Hispida.*

(6) The further development of the initials for the hypocotyl-radicle and root cap is precisely described. Detailed comparison is, however, unprofitable since *Myosotis Hispida* and Nolana appear to be fundamentally dissimilar. (cf. para 3 above).

(7) In *Myosotis Hispida* the suspensor consists of 2 to 3 cells, whereas in *Nolana Prostrata* and *Nolana Atriplicifolia* a long filamentous suspensor consists of 4 to 9 cells.

The above comparison shows clearly that after the formation of the 4-celled pro-embryo the law which governs the subsequent divisions in the formation of the embryo proper in *Myosotis Hispida* has no similarity with the law adopted in the development of embryo of these two Nolana species.

COMPARISON WITH CONVOLVULACEÆ.

Gertrude Macpherson (11) observed the following sequence in the development of the embryos of Dodder and Morning Glory :—" The first division of the fertilised egg in *Convolvulus Sepium* was transverse and resulted in the formation of an embryo very similar to that of *Cuscuta Gronovii.* The 4 and 8-celled stages were elongated and somewhat irregular in form, and much the same as in *Cuscuta*, but never exhibited the pronounced urnlike form. In stages of more than eight cells the embryo of *Convolvulus* is spherical in form, with a rather pronounced dermatogen in the majority of cases........................ In these advanced stages there is a very large suspensor consisting of large, very vacuolated, uninucleate cells, which completely fill the micropylar end of the sac and force the embryo well into the sac. The enormous development of the suspensor is much more rapid than that of the rest of the embryo, from which its separation is not always definite."

These facts have here been confirmed in the species, *Convolvulus Tricolor*. It has been said that in habit *Nolana Prostrata* approaches the Convolvulaceæ and particularly *Convolvulus Tricolor*. So the embryo formation of this species has here been specially compared with those of *Nolana Prostrata* and *Nolana Atriplicifolia* :—

(1) The very young stages of the embryo of *Convolvulus Tricolor* could not be handled for dissecting out, so the development of 4 and 8-celled stages has not been observed. But according to Miss Macpherson (11) these 4 and 8-celled stages of an allied species, viz. *Convolvulus sepium*, are elongated and somewhat irregular in form, whereas in these two Nolana species no such irregularity is observed. So even the early formation of 4-celled pro-embryo in the Convolvulaceæ has no resemblance to that of *Nolana Prostrata* and *Nolana Atriplicifolia*.

(2) The middle stages of the embryo of *Convolvulus Tricolor* have been easily dissected out as a whole. A characteristic of the embryo is the early development of chlorophyll pigments, so that the green colour helps to distinguish it from the surrounding tissue. After the 8-celled stage (cf. figure of a dissected out embryo CT 48 drawn under high power) a mass of irregular cells is formed in the pro-embryo. Till later the suspensor is not clearly differentiated. But in *Nolana Prostrata* and *Nolana Atriplicifolia* the embryo-forming apical cell is well differentiated early in the two-celled stage from the basal suspensor-producing one.

(3) During further developmental stages the embryo proper looks spherical in form and dermatogen is well marked (cf. figures from sectioned embryos CT 42, CT 43, CT 44, CT 45, CT 46, and figures of dissected out embryos CT 47, CT 49, CT 50, CT 51 drawn under high power and CT 52, CT 53 drawn under low power.) The law followed in the embryo formation of *Nolana Prostrata* and *Nolana Atriplicifolia* is not at all applicable to the formation of embryo of Convolvulaceæ.

(4) Massive tissue of suspensor, which acts as a haustorium, develops at first more vigorously than the tissue of the embryo (cf. figures CT 42, CT 43, CT 44, CT 45, CT 47, CT 49, CT 50, CT 51, CT 52, CT 53). These suspensor cells become larger in size and more vacuolated than the cells of the embryo (cf. figures CT 42, CT 43, CT 44, CT 45, CT 46). The massive suspensor is clearly seen also in the later stages (cf. figs. CT 46, CT 53, CT 54, CT 55, CT 56). This peculiar way of massive development of suspensor and embryo in Convolvulaceæ has no similarity with those of the two species, *Nolana Prostrata* and *Nolana Atriplicifolia* and as such Nolanaceæ, it appears, have no affinity with Convolvulaceæ.

Cytology.

Campin (4) was the first to study the cytology of *Nolana Atriplicifolia* and *N. Prostrata* and recorded the haploid chromosome number in both the spp. to be 12, and declared that the cytology of the two species is similar.

Whyte (20) in 1929 by his even a more thorough cytological study of these two parent spp. came to the same conclusion. Miss Saunders (13) in 1930 confirmed the same haploid chromosome numbers for both the species.

The diploid chromosome number of 24 has been counted in the young embryo of *Nolana Atriplicifolia* and in the integument of the ovule (Fig. NP 57) in *Nolana Prostrata*. The triple number of chromosomes, viz :—36, in the endospermic nuclei of *N. Prostrata* has also been counted (Fig. NP 58).

Saunders' (13) " Discussion on the influence of the cytoplasm " of these two species when crossed, may, with profit, be summarised here :—

" Prostrata ♀ × Atriplicifolia ♂ usually fails, but succeeds occasionally. The crossbreds differ from both parents in the colour pattern of the flower. The pollen is largely " bad ". As a rule, the last flowers of the season alone set seed.

These F_1 crossbreds pollinated with *Prostrata* yielded little seed, but the back-cross with Prostrata as ♀ proved somewhat less sterile. With *Atriplicifolia* the back-cross either way scarcely ever gave a seed.

The reciprocal mating, *Atriplicifolia* ♀ × *Prostrata* ♂, invariably failed. Nevertheless, there is strong indirect evidence that in nature this union is sometimes fertile, yielding plants indistinguishable in flower from *Atriplicifolia*.

Putative natural hybrids of this origin were indistinguishable in flower from the mother form. When selfed or pollinated with *Atriplicifolia* they yielded only *Atriplicifolia* through successive generations. When crossed back on *Prostrata* ♀ they gave *Prostrata* and the reciprocal crossbred in the ratio 1 : 1.

The above facts together with the occurrence of " reversed dominance " of certain characters in lateral generations suggest that inheritance may here be determined partly by the cytoplasm."

This unusual genetical behaviour made it appear of some interest to study reciprocal hybrids of these two Nolana species with a view to find out whether or not there is any abnormality in their chromosomes. (My very grateful thanks are due to Miss E. R. Saunders for offering the material.)

In the reciprocal hybrids *Nolana Prostrata* ♀ × *Nolana Atriplici-folia* ♂ only the young ovules have been worked out. In some of these hybrids the development of normal type of 4 megaspores have been observed. The figs. NH 59, NP ♀ × NA ♂, shows the stage after the formation of 4 megaspores. Three of them are crushed and one enlarged. Chromosomes in these plants have not been counted. But in the hybrid *Atriplicifolia* ♀ × *Prostrata* ♂ (fig. NH 63, NA ♀ × NP ♂) on the wall of megasporangia and microsporangia 24 diploid chromosomes have been counted. In the root-tip of *Nolana Paradoxa* (the "Putative natural hybrid") 24 chromosomes are found (fig. N. para 62). The contribution to Miss Saunder's work is therefore rather negative. All forms, both parents and hydrids, have the same number of chromosomes. The available material has unfortunately been too scanty for the elucidation of the chromosome behaviour throughout the whole life cycle but such evidence as has been obtained gives no suggestion of gross abnormality.

It is, however, interesting to note as a further evidence of relation-ships between Nolanaceæ and Solanaceæ that the basic number of chromosomes is 12 in *N. Atriplicifolia* and *N. Prostrata*. Tischler (17) found the same haploid number in many of the species belong-ing to Solanaceæ. In the present work, in the root-tip of *Solanum Dulcamara* presence of 24 diploid chromosomes has been confirmed. Figures of chromosomes in the root-tip of *N. Paradoxa* (fig. N. para 62) and in the ovary wall of the hybrid plant (Fig. NH 63, NA ♀ × NP ♂) show resemblance to *Solanum Dulcamara* (fig. Sol. Dul. 61) in their general form and in the possession of a pair of chromosomes with satellites. On the other hand only in a few species of Boraginaceæ, worked out by Stray (17) in 1930, the haploid chromosome number 12 is found. But the commonest numbers of chromosomes in Boraginaceæ are 7 or 8 or multiples of 7 or 8. In this work in the root-tip of the species *Symphytum Officinale* in Boraginaceæ, 42 chromosomes have been counted (Fig. B. S. 60). But in no species of Convolvulaceæ has the haploid number 12 been described (17). From the above available cytological data in Solanaceæ, Boraginaceæ, Convolvulaceæ and Nolanaceæ it seems Nolana has nearest relation-ship with Solanaceæ.

Summary.

From the systematic position of the Nolanaceæ as described by different authorities this group has possibilities of affinity with the three groups ;—(1) Solanaceæ, (2) Boraginaceæ and (3) Convolvulaceæ. So the aim of the work was to elucidate to which of these three groups *Nolana Prostrata* and *Nolana Atriplicifolia* and as a matter of that the group Nolanaceæ have the nearest affinity.

The following facts have been observed in the embryo-development of the two Nolana species :—

(1) From the two-celled pro-embryo the apical cell segments transversely to give rise to 4-celled stage. The apical cell divides before the basal one. It appears the apical cell only takes part in the formation of the embryo proper and the basal one produces suspensor only. The pro-embryo seems to be a long 6 to 8-celled filamentous one.

(2) The three apical cells I, II, III take part in the formation of the embryo as follows :—

<div style="margin-left:3em">
I gives rise to cotyledon and stem-apex

II   „   „   „ hypocotyl and radicle

III   „   „   „ middle of the root-cap.
</div>

(3) In the early segmentation of tissues in the octant stage (i.e. 4 cells in the tier I and 4 cells in the tier II) the dermatogen is cut off by periclinal divisions.

(4) After the differentiation of dermatogen in both the tiers, I and II, two layers are formed in the penultimate layer II to separate hypocotyl from the radicle. During further growth of hypocotyl and radicle, sometimes in radicle region the central cylinder of the root is early separated from the cortex of the root by oblique division. Otherwise the central cyinder of the root is differentiated from the cortex of the root only after the formation of a fairly massive tissue.

(5) In the cotyledonary tier I two layers are formed early· There appears to be nothing remarkable in the further growth of the cotyledons. They are curved in the ripe seed. Stem-apex is presumably formed from the apical tier I, but does not appear to develop till the seed germinates.

(6) The suspensor consists of a long filamentous structure of 4 to 9 cells. The basal one is haustorial.

(7) Some abnormal embryos are described.

(8) The haploid chromosome number in both the species is 12.

(9) Similarity in chromosome morphology in *N. Paradoxa* and *S. Dulcamara* noted.

(10) In reciprocal hybrids between the two spp. of Nolana there is no evidence of gross irregularity in chromosome behaviour.

(11) From a study of the embryo development and cytology in Nolanaceæ, Solanaceæ, Boraginaceæ and Convolvulaceæ the conclusion is drawn that *Nolana Prostrata* and *Nolana Atriplicifolia* possess a type of embryo development of their own although the fundamental law in embryo-segmentation is like that of Solanaceæ. They have no affinity with the Boraginaceæ and Convolvulaceæ.

In conclusion I take this opportunity of expressing my deep gratitude to Dr. Manton who watched the progress of my work with keen interest and helped me, whenever necessary, with her very useful and kind advice. I am very grateful also to Prof. Drummond under whose general supervision and guidance my work was carried on. My sincere thanks are also due to the Curators of the Herbarium in the Kew Gardens and the Cambridge Botanic Garden, who kindly permitted me to observe there some very interesting specimens required in connection with my research work.

Bibliography.

1. BENTHAM, G ET J. D. HOOKER.—Genera Plantarum Vol. II. Pars prima, sistens ordines LXXXIII ad LXXXVIII p. 1 ad 533, edita fuit mense Aprili, 1873.

2. BLAKESLEE, A. F. AND JOHN BELLING.—The Reduction Division in Haploid, Diploid, Triploid and Tetraploid Daturas.—Proceedings of the National Academy of Sciences of the U.S.A. Volume 9, 1923.

3. BROWN, M. M.—The development of the embryosac and the embryo in Phaseolus Vulgaris. Bull. Torr. Bot. Club. 44. 535–544. Taf. 25–26, 1917.

4. CAMPIN, M. G.—A cytological study of Pollen development in Nolana. New Phytol., 24, 16–23, Taf. I, II, 1925.

5. CANDOLLE De.—Prodromus—Systematis Naturalis Regni Vegetabilis. Pars. XIII, Sect. 1, 1852.

6. COULTER AND CHAMBERLAIN.—Morphology of Angiosperms, 1903.

7. ENGLER, A. AND K. PRANTL.—Die Naturlichen Pflanzenfamilien. IV. Teil. 1 and 2 Abteilung. 1891.

8. HUTCHINSON, J.—The families of flowering plants. I. Dicotyledons, 1926.

9. MACPHERSON, GERTRUDE E.—Comparison of development in Dodder and Morning glory. Bot. Gaz. 71 392–398. Taf. 25–27. 1921.

10. MANTON, I.—Introduction to the general cytology of the Cruciferæ.—Ann. Bot. Vol. XLVI. No. CLXXXI. 1932.

11. MIERS, JOHN.—Illustrations of South American Plants. Vol. II. 1849 to 1857.

12. RENDLE, A. B.—The Classification of flowering plants. Vol. II. Dicotyledons. 1925.

13. SAUNDERS, E. R.—Discussion on the influence of cytoplasm.—The Heredity Mechanism in Nolana. Fifth International Botanical Congress, Cambridge—Report of proceedings By F. T. Brooks and T. F. Chipp. 1930.

14. SCHNARF, K.—Handbuch der Pflanzenanatomie. II Abteilung 2 Teil : Archegoniaten. Embryologie der Angiospermen. 1928.

15. SHARP, L. W.—An Introduction to Cytology. McGraw Hill Book Company, 1926.

16. SOUEGES.—Recherches sur l' embryogenie des Solanacees. Bull. Soc. Bot. France, 69, 163–178, 236–241, 352–365, 555–585. 1922 a.

17. SOUEGES.—Development de l' embryon chez le Myosotis hispida. Bull. Soc. bot. France, 70, 385–401. 1923.

18. TISCHLER.—Pflanzliche chromosomen-zahlen Tabulae Biologicæ Periodicæ Vol. VII. 1931.

19. WARMING, E.—A Handbook of systematic Botany. Translated and edited by M. C. Potter, M.A., F.L.S. 1895.

20. WETTSTEIN, R. V.—Handbuch der systematischen Botanik. 1911.

21. WHYTE, R. O.—Chromosome Studies II. Interspecific Hybrids in the Genus Nolana. New Phytol. 28. 1929.

Explanation of Plates.

PLATE I. FIGS. 1-25.

(Figures reduced to magnification 240 from the original figures magnified 960. N P=Nolana Prostrata, N A=Nolana Atriplicifolia, Der=Dermatogen. h=hypocotyl, r=radicle, sus=suspensor.)

N P 1—Two-celled pro-embryo. Apical cell C A, basal cell C B.

N A 2—Three adjoining sections of the same two-celled pro-embryo. Apical cell C A in mitosis.

N A 3—Four-celled pro-embryo, viz :—l, l', m & ci.

N A 4—Four-celled pro-embryo l, l', m & ci, the penultimate one is in mitosis.

N P 5—6-celled filamentous pro-embryo. Probable 4 terminal cells derived from l & l'. Three terminal tiers named I, II, III taking part in the formation of embryo proper.

N A 6—Beginning of embryo proper. Two end tiers I & II enlarged and divided longitudinally.

N P 7—As N A 6.

N A   8—Penultimate II divided longitudinally.

N P   9—Quadrant stage of the embryo (2 cells in the tier I and 2 in II)

N P 10—4 cells formed in the penultimate tier.

N P 11—Octant stage of the embryo (4 cells in the tier I and 4 cells in II)

N A 12—After octant stage of the embryo left hand cell in the penultimate one II is in mitosis periclinal to separate off dermatogen.

N P 13—Dermatogen differentiated in the penultimate II and also in the right hand cell of the tier I by periclinal division.

N P 14—Dermatogen differentiated in both I & II. III tier divided. Sus—6 cells.

N A 15—As N P 14.

N P 16—As N P 14 but dermatogen in the cotyledonary tier I divided anticlinally. The 2 cells in III are in mitosis.

N A 17—Right hand inner cell in the tier II divided into two layers to separate hypocotyl, viz:—h from radicle, i.e., r and the left hand one is in mitosis. III tier still undivided. Sus 5.

N A 18—Same stage as N A 17, tier III divided.

N A 19—Two tiers formed in the tier II and two tiers in the right hand cell in the tier I.   III tier is in division. Sus 6 cells.

N A 20a & 20b—Two adjoining sections of the same embryo to show longitudinal division of dermatogen of tier II but transverse division of central cells.

N A 21—The layers, viz:—h and r in the tier II and two superposed layers in the right hand side of the tier I. III tier still inactive. Long sus—9 cells, broad and narrow. Basal haustorial cell divided obliquely.

N P 22—Two superposed layers in both tiers I and II. III tier divided vertically. Long sus—9 cells broad and narrow as in N A 21.

N P 23—Two layers formed in both tiers I and II and middle cell in the tier r divided vertically. III tier still undivided. Sus 5 cells.

N A 24—Two layers r and h in the tier II and further vertical division in the middle cells of r and in the right hand middle one in h.   2 superposed layers also in cotyledon tier I.

N A 25—Two superposed layers formed in both I and II and the left hand cell in the tier r by a curved wall to differentiate central cylinder of the root from its cortex.

PLATE II. FIGS. 26-36.

(Fig. N A 35 of a dissected out embryo and N A 36 a sectioned embryo of a ripe seed reduced to magnification 24 from the original figures magnified 97. All other figures in this plate reduced to magnification 255 from the original figures magnified 960)

N A 26—Both longitudinal and transverse divisions in the central cells of layers h and r (central cylinder and cortex of the root are not differentiated early as in N A 25 in the plate I). III and IV tiers divided vertically sus 5, basal haustorial cell divided obliquely.

N P 27—Further advanced stage. Central cylinder and cortex of the root still undifferentiated. III in 8 cells formed by nearly vertical divisions (4 visible in section) Sus 5 longer in size.

N A 28—Tier III divided transversely.

N A 29—Further advanced stage. Cells in tier III obliquely divided. One cell in mitosis in the cotyledonary tier shows 24 chromosomes.

N A 30—Central cylinder and cortex of the root differentiated in the tier r. 4 cells in III obliquely divided and tier IV segmented vertically. Sus 5, basal haustorial cell divided obliquely.

N P 31—Same stage as N A 30. IV tier undivided.

N A 32—Two adjoining sections of the same embryo. Advanced stage but III tier consists of 4 cells.

N A 33—Whole root-cap and its external layer of the middle part derived from the tier III.

N P 34—IV tier divided vertically.

N A 35—Dissected entire embryo. 2 cotyledons bulge out.

N A 36—Section from ripe seed. Cotyledons curved. Still stem-apex not formed.

PLATE III. FIGS. 37-56.

(Figures N P 37, N A 38, N A 39, N P 40, N A 41 reduced to magnification 260 from the original figures magnified 960. Figs. C T 42-C T 46 reduced to 155 from original figures magnified 520 drawn under ordinary high power. C T 47 to 51 figures of entire dissected embryos same reduced magnification to 180. Figs. C T 51 to 56 reduced to magnification 27 from the original figures drawn under low C T = Convolvulus Tricolor.

N P 37—Long filamentous pro-embryo. Apical cell abnormally. divided.

N A 38—IV tier early divided.

N A 39—Middle cells in the tier r first divided transversely. Tier III divided transversely.

N P 40—Vertical division in the tier III, tier IV divided whereas tier I slow in divisions.

N P 41—Tier I slow in division, other tiers seem to divide without a definite order.

C T 42—C T 46—Different stages of developing embryos of Convolvulus Tricolor (microtome sections). Embryo proper spherical in form. Dermatogen differentiated, massive suspensor divided more vigorously, cells vacuolated, larger in size.

C T 47—C T 56 Different stages of dissecting out entire embryos of Convolvulus Tricolor.

PLATE IV. FIGS. 57-63.

(All figures of chromosomes magnified 2,600 same as from original figures. Fig. N H 59, N P ♀ × N A ♂ is in same magnification 960 as in original figure.

Ch=chromosome, N H=Nolana Hybrid.

N P ♀ = Nolana Prostrata Female, N A ♂ = Nolana Atriplicifolia male.

Sol. Dul. = Solanum Dulcamara. B. S. = (Boraginaceæ) Symphytum Officinale.

N A ♀ = Nolana Atriplicifolia Female, N P ♂ = Nolana Prostrata male.

N P 57 —24 diploid chromosomes in integument.

N P 58 —36 triploid chromosomes in Endosperm.

N H 59 (N P ♀ × N A ♂) —4 megaspores within nucellus. Three megaspores crushed (black ones).

B. S. 60 —42 chromosomes in the root-tip.

Sol. Dul. 61 —24 chromosomes in the root-tip.

N. Para 62 —24 chromosomes in the root-tip.

N H 63 (N A ♀ × N P ♂) somes in ovary wall.
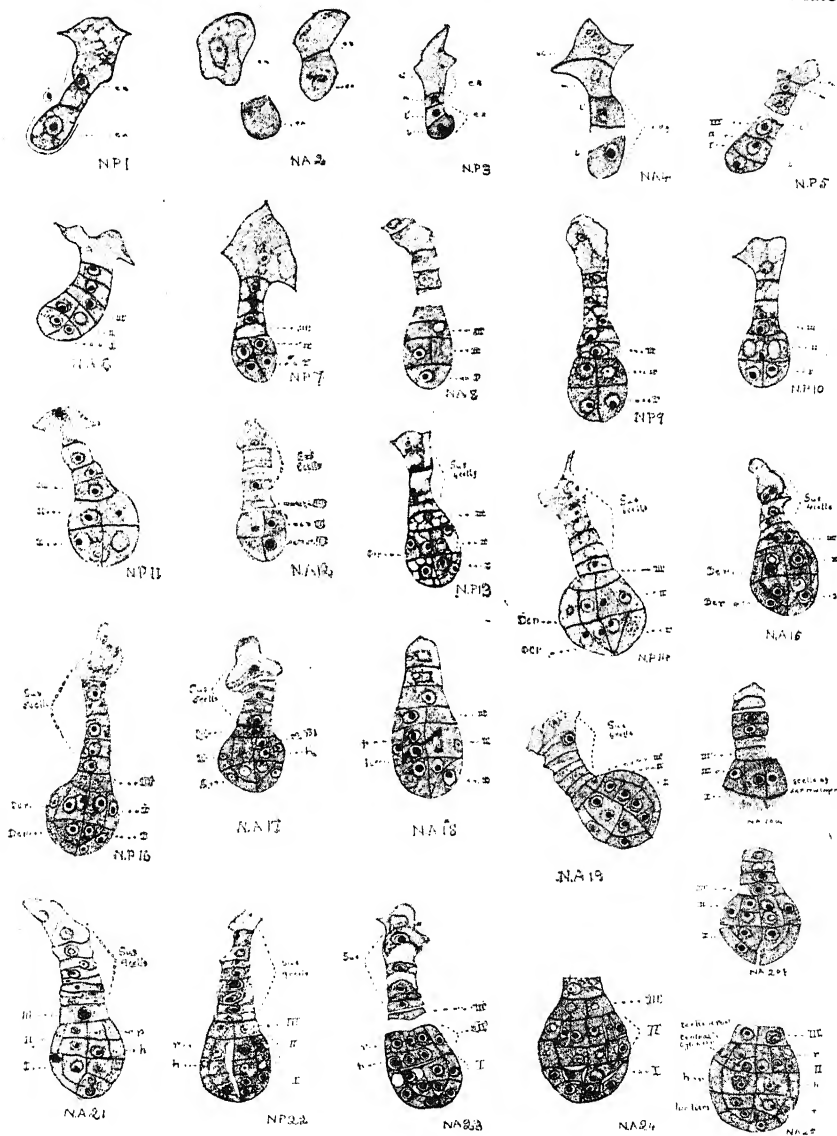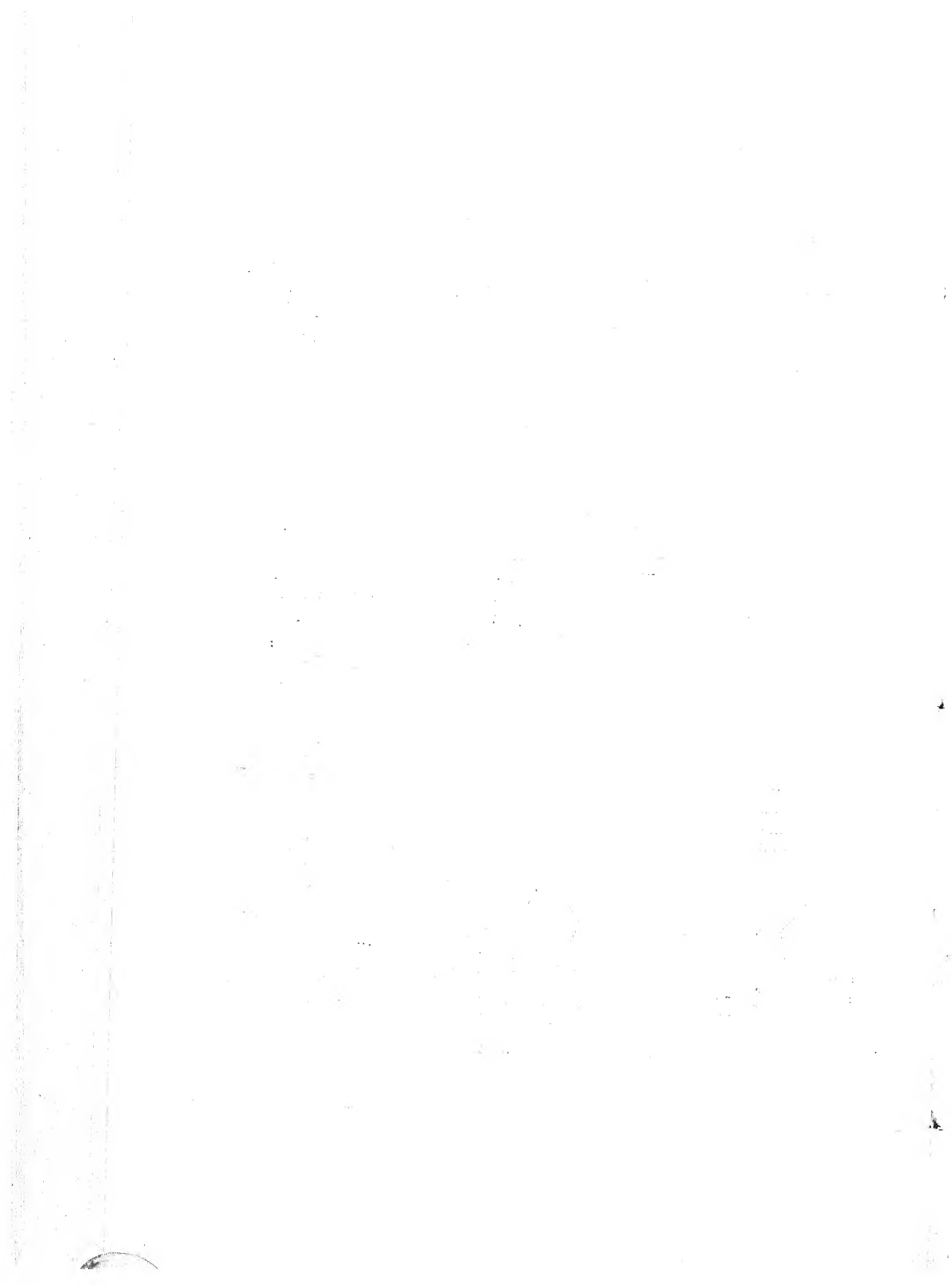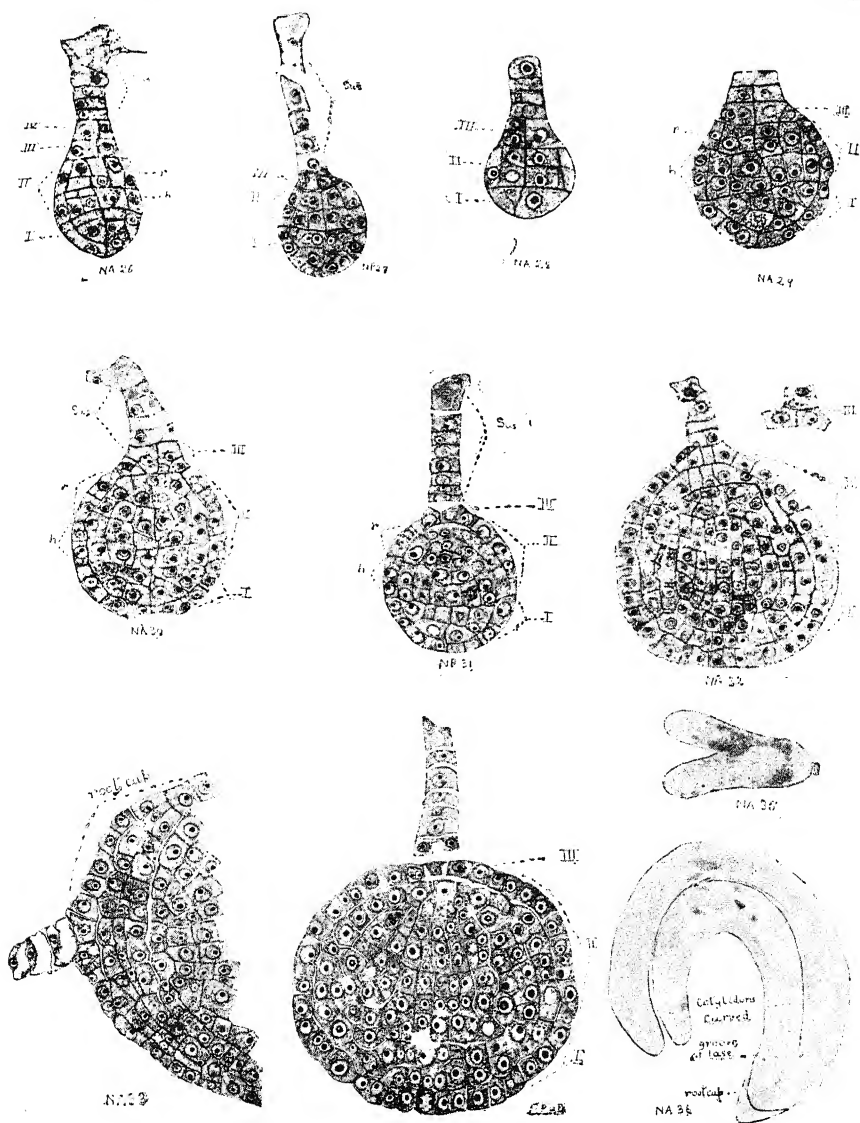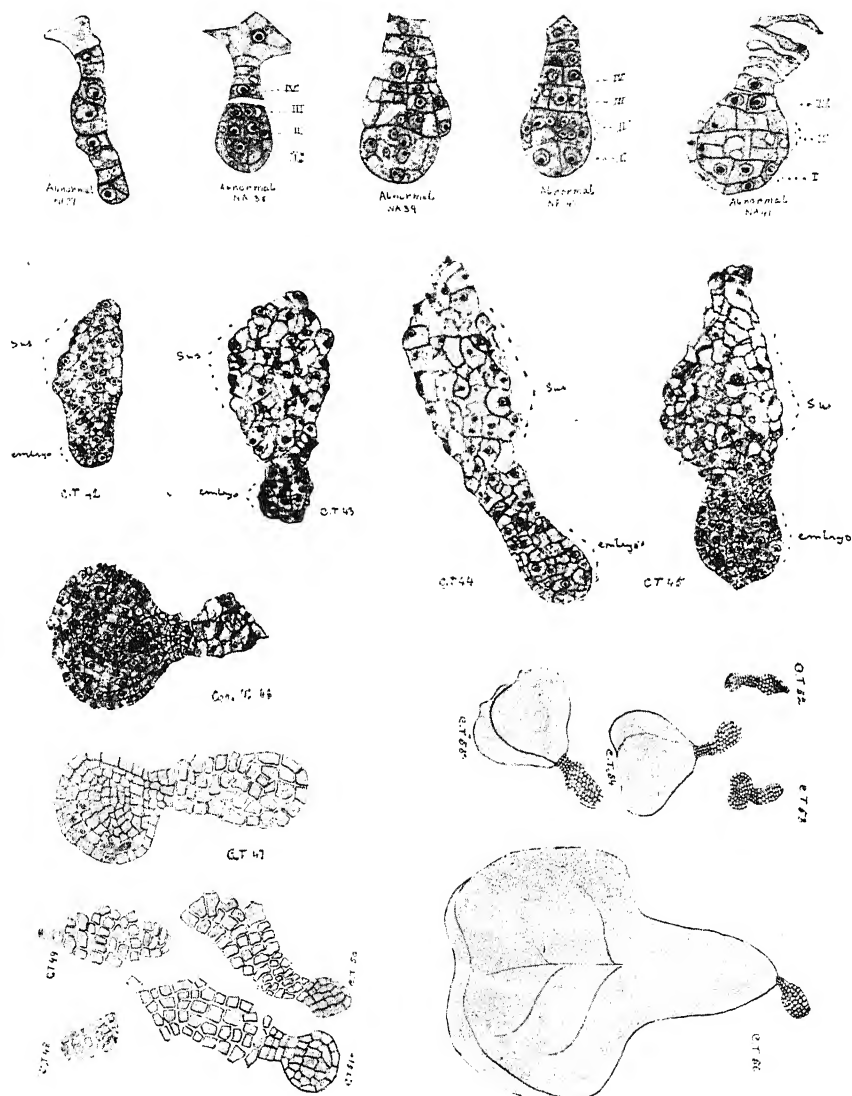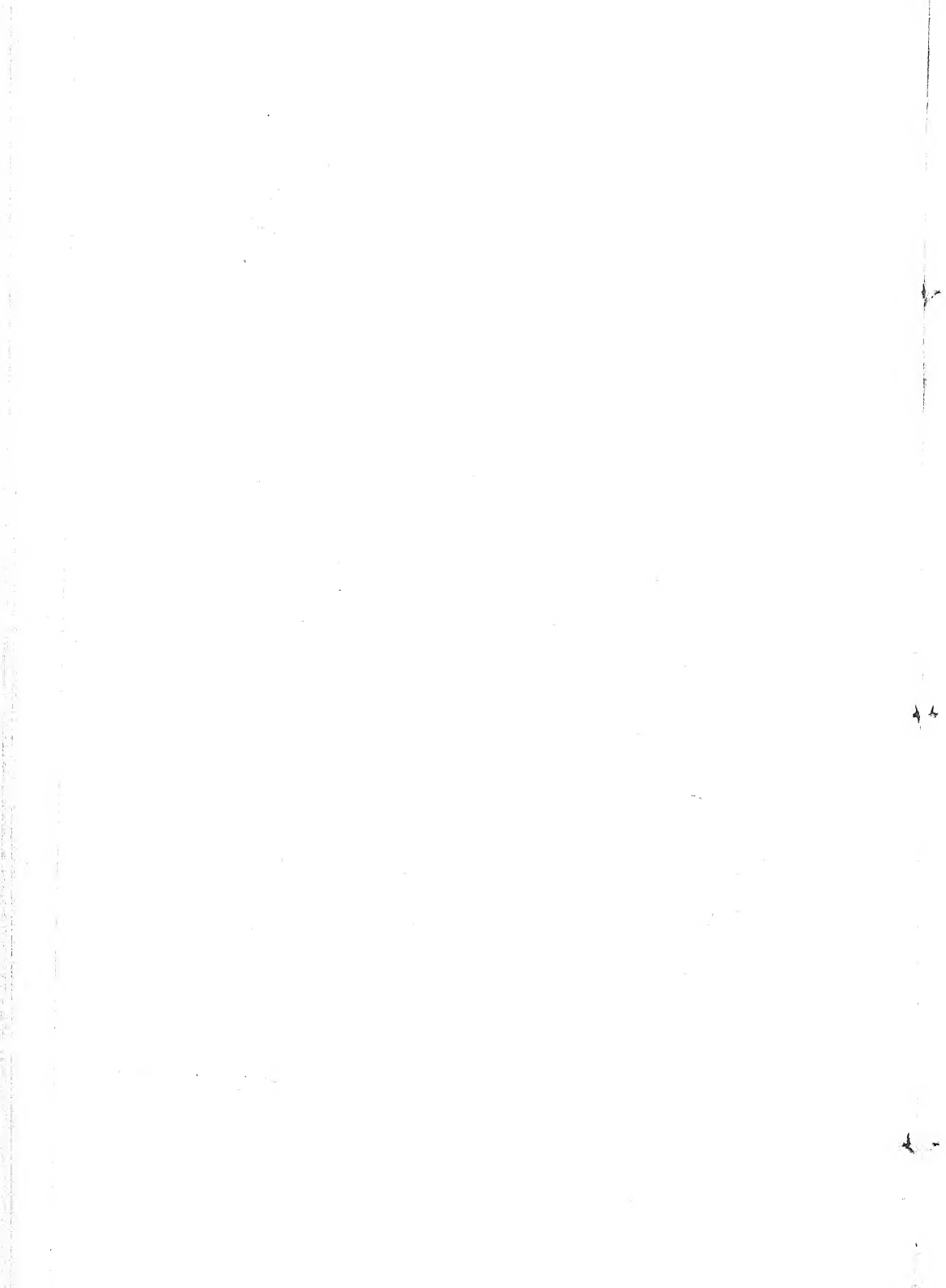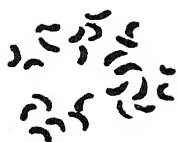
ch. 24 (Integument)
N.P 57

ch. 36 (endosperm)
N P 58
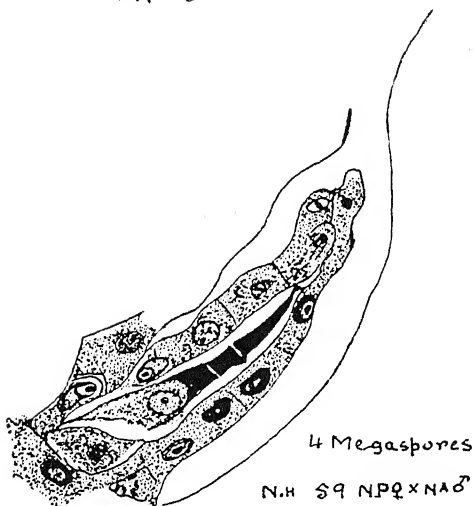
4 Megaspores
N.H 59 NP♀ × NA♂

ch. 42.
B.S. 60

ch. 24.
Sol.Dul 61

ch. 24
N. Para 62

ch.24 (ovary wall)
N.H.63 NA♀ × NP♂

A CONTRIBUTION TO OUR KNOWLEDGE OF INDIAN COPROPHILOUS FUNGI

BY

NAZEER AHMAD MAHJU,

Botany Department, Panjab University, Lahore.

Contents.

Introduction.

The Coprophilous Fungi, i.e., fungi growing on dungs, have not been systematically studied in this country. If carefully studied many interesting forms will no doubt be added to the list of our present fungal flora.

Important contributions on dung flora have so far been mostly made from Europe, and among others the work of Hansen in Denmark ; Boudier and Cronan in France; Coemans and Marchal in Belgium; Spegazzini in Italy ; Heimeral and Zukal in Austria; Winter and Zopf in Germany ; Karsten in Finland and Chelchowsky in Poland deserves special mention.

The members of all the four great divisions of fungi are fairly represented in the dung. The largest number of individual species of fungi are found in the dungs of herbivorous animals. According to Saccardo, the dungs of herbivora, carnivora and reptilia bring forth seven hundred and eight, forty-five, and four species respectively. Collectively there are nearly seven hundred and fifty-seven species in one hundred and eighty-seven genera. Other fungi especially. those of Hyphomycetes do not confine themselves to the dung, but may also grow on decaying vegetable substances. As the majority of coprophilous fungi grow on herbivorous dung, the general distribution will be influenced by the relative number of such animals in a given region.

In this paper it has been possible to study the dungs of 6 animals thoroughly. These are, rabbit, sambhar, horse, goat, buffalo and sheep.

The dungs were collected fresh from the Zoological Garden, Lahore. These dungs were placed in sterilised dishes which were placed under the bell-jars on glass plates and sealed with vaseline to prevent contamination.

For the study of the Hyphomycetes temporary mounts of the material were made in Glacial Acetic Acid, because the conidia fall away in water giving no clue to its identity. For the permanent mounts material was stained in Alcoholic Eosin and mounted in Balsam.

Generally for staining the material, Alcoholic Eosin was largely used and proved very satisfactory. In Sordareæ the stained portions were only the asci and the mucilaginous appendages of the spores. Chætomium spirale and Magnusia nitida were mounted unstained in Glycerine.

The dungs were examined every day under the binocular which easily revealed the birth of every new species. The petri-dish was always kept covered with glass plate while examining.

Altogether 29 different species in 21 genera have been recorded from these 6 different dungs.

The writer expresses his indebtedness to Dr. H. Chaudhuri of the Panjab University for general guidance and helpful advice throughout the period of the investigation and in the preparation of the manuscript.

General Characters of the Coprophilous Fungi.

As Coprophilous fungi do not form a concrete group, they cannot be treated from a comparative point of view, nevertheless some structural features claim attention. The most typical of the ascigerous coprophilous fungi are those included in Sordareæ and Ascoboleæ. Spores in both of these groups are comparatively large and deeply coloured. In the genus Ascobolus spores are hyaline when young but become brown when reaching maturity. Similarly in Sordareæ the spores pass a series of colours to reach the final opaque stage.

The spores in the case of Ascobolus are surrounded with mucilage in some species, whereas in Sordaria macrospora the mucilage forms a hyaline refractive belt. In Sordaria curvula a hyaline appendage is attached to the sides of the spores. It has been observed that when the spores are fully mature, the appendage disappears, while in the young condition it is very prominent.

In many of the dung borne fungi, the sporangium or spores are ejected at a long distance. In the case of Sordareæ spores are ejected in a mass to a considerable distance. Ascospores of many of these coprophilous fungi are difficult to germinate unless they pass through the alimentary canal of animals.

Examination of the dung kept in the laboratory shows, that various species of fungi do not confine themselves to any specific habitat. It is obvious, therefore, that in many cases at least no character of

specific value can be attached to the occurrence of a species on the dung of any particular animal.

The perithecia in Sordareæ are immersed to various degrees in different dungs. This probably, as Massee remarks, depends upon the hard or soft texture of the sub-stratum.

It has been found that the number of individual plants belonging to a particular species differ in different dungs, for example, Sordaria macrospora and Ascobolus viridis are abundant in sheep and buffalo dungs respectively, whereas this is not the case in the other dungs where both of these species are present.

Regarding sequence of appearance of the different fungi, it has been noticed that Phycomycetes, usually heralded with Pilolobus, followed by species of Mucor. Next various members of Hyphomycetes appear and then the Basidiomycetes. The Ascigerous fungi are usually the last in the sequence to appear.

Table of Fungi.

The following table shows the species which appeared in various dungs during the course of 6 months (December to May).

No.	Rabbit dung	Sambhar dung	Horse dung	Goat dung	Buffalo dung	Sheep dung
1.	...	Mucor mucedo.	M. mucedo	...	M. mucedo	...
2.	...	Pilobolus longipes.	P. Longipes.	...	P. Longipes.	...
3.	...	...	...	...	Pilobolus minutus	...
4.	...	...	Pilobolus crystallinus.	...	P. crystallinus.	..
5.	...	...	...	Myxotrichum charatum.	...	...
6.	...	...	...	...	...	Aspergillus flavus.
7.	...	...	...	Myxotrichum aeruginosum.	...	...
8.	...	...	...	Magnusia nitida.	...	...
9.	...	Sordaria macrospora.	S. macrospora	S. macrospora	S. macrospora.	S. macrospora.
10.	...	Sordaria curvula.	S. curvula.	...	...	...

Table of Fungi—(Contd.)

No	Rabbit dung	Sambhar dung	Horse dung	Goat dung	Buffalo dung	Sheep dung
11.	...	...	...	Chætomium spirale.	...	...
12.	...	...	...	Sporomiella nigropurpu-rea.	...	...
13.	...	...	...	Pezolepis sp.	...	...
14.	Ascobolus viridis.	...	...	Ascobolus viridis.	A. viridis.	...
15.	...	...	...	Lasiobolus hirtellus.	...	...
16.	...	...	...	Lachnella fraxinicola.	...	...
17.	...	...	...	Lachnella albido-fusco.	...	...
18.	...	...	Coprinus niveus.	...	...	...
19.	...	Coprinus papillatus	...	...	...	...
20.	...	...	Coprinus radiatus	...	...	...
21.	Coprinus ephemerus.	...	...	...	...	...
22.	...	...	Bolbitus vitellinus.	...	...	...
23.	Dendrostil-bella byssina.	...	...	...	...	...
24.	...	...	...	...	Silicipodium sanguineum.	...
25.	...	...	Oedocepha-lum glomeru-losum	Oe. glomerulo-sum.	Oe. glomerulo-sum.	...
26.	Arthrobo-trys superba.	...	...	...	...	...
27.	Stysanus stemonitis	...	...	...	...	S. Stemonitis
28.	Torula convoluta.	...	...	...	...	...
29.	...	...	...	...	...	Isaria brachiata.
Total No.	6	5	10	12	8	4

Descriptions.

1. *Mucor mucedo*. (Link), Eng. and Prant. Nat. Pflan. Fam. Teil I. I. ABT., p. 124. Fig. 106; Sacc. Syll. Fung. Vol. VII, p. 191.

Sterile hypheae creeping, branched, septate; sporangiophores erect, unbranched, smooth, aseptate, 1–4 cm. high 5·4—12 μ thick; sporangium terminal, globose, yellow when young becoming greyish with age, 5·4—12 μ in diameter; columella cylindrical, club-shaped, 6–21·5 × 40—148 μ (from various specimens in various grades of development); spores broadly elliptical, hyaline, 4·5—17 × 2·5—7 μ.

Habit :—Buffalo, Horse and Sambhar dung.

2. *Pilobolus longipes* (Van Tiegh.); Sacc. Syll. Fung., Vol. 7, p. 185.

Sporangiophore erect, elongated, 1·8 to 2 mm. high, apice globose, base bulbous, basal mycelium long, extending, sub-cylindrical, yellow; sporangium 310 to 400 μ in diam. Colour uniform, dark; spores elliposid to sub-sphaeroid, epispore rough, thick, cartilage-like, 9 to 12 × 8 to 11 μ.

In the young condition the plant is quite yellow, especially the sporangium which becomes dark with age.

Habit :—Buffalo Dung, Horse and Sambhar dung.

3. *Pilobolus minutus*. (Speg.) Sacc. Syll. Fung., Vol. 7, p. 186.

Sporangiophore superficial, gregarious, 2 to 2·5 mm. high, first filiform then swollen at the apex and elliptical at the base, hyaline; base, bulbous, tapering downwards, covered with minute drops of water; sporangium soft, greyish, 3 mm. in diam. ; spores elliptic to sphaeroid, hyaline, granular, 8 to 9 μ in diam.

Habit :—Horse and Buffalo dung.

4. *Pilobolus crystallinus*. (Wigg.) Sacc. Syll. Fung., Vol. 7, p. 185.

Sporangiophore slender, 4 to 4·5 mm. high, yellowish, bedewed, transparent, apice swollen, club-shaped; sporangium hemispherical, 310 μ in diam., black, cuticle verrucose, reticulate, columella bluish, contains numerous spores; spores of uniform shape, elliptic, epispore thin, 6·2 to 9 × 3·7 to 4·5 μ, light yellow.

Habit :—Horse and Buffalo dung.

5. *Aspergillus flavus*. (Link) Thom, C. & Church, M. B., Asperg., p. 199.

Sterile hypheae creeping, effused, dirty white; fertile hypheae aseptate, erect, 7·2 to 8 mm. high, 24·3 to 33·7 μ wide, attenuated towards the base, swollen in apex into a head; head 40 μ long and 35 μ wide, at first white then becoming more or less greenish yellow, sterigmata in one series, densely covered with chains of conidia, conidia minute, hyaline, elliptic with obtuse ends, 4·7 to 6·2 × 3 to 3·8 μ.

Habit :—Sheep dung.

6. *Myxotrichum charatum* (Kunz), Sacc. Syll. Fung., Vol. 4, p. 317.

Glomeruli gregarious, flask-shaped with a short neck 270 to 324 μ in diam., neck cellular; hypheae thick-walled, black, united, forming a lattice-like peridium, much longer than glomerulus, 18 to 607 × 5·4 μ smooth, uncinate at the apices, septate; asic minute, densely crowded into a yellowish mass, globose, octo-sporous; spores faintly yellow, oval, smooth 6·2 to 7·2 × 4·6 to 6 μ.

Habit:—Goat dung.

7. *Myxotrichum aeruginosum*, (Mont.), Sacc. Syll. Fung, Vol. 4, p. 319 (1886).

Glomeruli scattered, roundish flat, 172 μ in diam. blackish, superficial; hypheae dense, intricate, irregular, septate, thick-walled, united to form a network; appendages spinous, erect, tapering at the tip, 384 to 140 μ in length. Spores hyaline with a slight grey tinge, oval, smooth, 12·5 to 14 × 6·2 to 7·9 μ.

Habit:—Goat dung.

8. *Magnusia nitida.* (Sacc.) Sacc. Syll. Fung., i. 38 (1882).

Perithecia scattered or sub-gregarious, superficial, sub-globose, membranaceo-carbonaceous, black about 1 mm. in diam.; wall dense, parenchymatous, fragile, composed of small polygonal cells, diam. about 5 μ; appendages springing from the both sides of perithecium, from 6 to 20 in number, horizontally spreading somewhat rigid, dark brown, opaque about 675 to 767 × 4 μ, regularly circinate at the apex, asci oblong or oblong-pyriform, octosporous, very evanescent; spores broadly elliptical, acute at both ends 4 × 3 μ, smooth, at first hyaline becoming steel grey.

Habit:—Goat dung.

9. *Sordaria macrospora.* (Auersw.), Wint., Deutsch. Sordar. 79, Taf. VII, f. 4 (1873).

Perithecia black, gregarious or crowded, glabrous, semi-immersed, sometimes superficial, 1/3 to 1/2 mm. in height, basal part sub-globose, narrowed upwards into a short bluntly conical neck, perithecial wall parenchymatous, cells 14 μ, tapering below into an evident stalk, apex rounded octo-sporous. Spores yellowish green when young and black at maturity. Uniseriate broadly obovate or oblong, rounded at the apical end and minutely pointed at the basal end, 20 to 25 μ × 13 to 17 μ.

Habit:—Goat, Sambhar, Buffalo, Sheep and Horse dung.

10. *Sordaria curvula.* (De Bary), Wint., Deutsch. Sordar. 101, Taf. XI, f. 22 (1873); Sacc. Syll. Fung i. 233 (1882).

Perithecia blackish, somewhat transparent, gregarious, 445 μ long immersed to the base of the neck, basal part ovate globose, narrowed

upwards into long obtuse neck, wall parenchymatous, asci cylindrical, shortly pedicellate, broadly rounded upwards, octo-sporous, 162 to 175 × 27 to 29 μ; spores ellipsoid, 25 to 29·5 × 14 to 17 μ, with a hyaline cylindrical basal appendage, 18·7 × 4·7 μ; a little shorter than the spores.

Habit :—Horse and Sambhar dung.

11. *Chætomium spirale.* (Zopf.), Eng. and Prant. Nat. Pflan. Fam. Teil, I. I. ABT, p. 388, Fig. F, p. 389.

Perethecia scattered or sub-gregarious, oval or elliptical 148 to 189 μ high, with numerous crowded, mostly spirally wound hair, which are fuscus and multi-septate, often become very rough, lateral hair spreading, simple, straight; wall of the perethecium cellular, light grey in colour; spores bi-seriate, sub-globose, 5·6 to 6·2 × 5·6 to 6 μ.

Habit :—Goat and Sheep dung.

12. *Sporomiella nigropurpurea.* (Ell. and Everh.), Sacc. Syll. Fung. II, 330.

Perethecia soft, carnose, aggregated into groups, forming more or less extended continuous black patches, 175 to 189 μ; asci numerous, 84 to 103 × 9 to 12 μ, narrowly cylindrical, often flexuose, narrowed below into a stalk, octosporous; spores bi-seriate, sub-cylindrical, 28 to 31 × 5·6 μ, surrounded with a narrow layer of mucilage, nearly straight, tetra-merous, median cells sub-globose, terminal cells sub-ovate; perithecial walls parenchymatous.

Habit :—Goat dung.

13. *Pezolepis* sp. (Syd.), Clem. and Shear. Gen. Fung., p. 114 (1931) (Ann. myc. 23 : 408. ill. (1925).

Apothecia gregarious, variable in size, elongated, disc flat, superficial, dark to dark brown, ·6 to ·9 mm. width × ·9 mm. height, glabrous, superficial and leathery; asci and paraphyses enclosed in the exciple, numerous, arranged in vertical rows, clavate, with a stalk-like base, 135 to 189 μ in length and 13·5 to 20 μ in breadth, octo-sporous; spores oval, hyaline, arranged in a single row, smooth, 13·5, to 18 μ in length, 11 to 13 μ in breadth; paraphyses hyaline filiform, septate surrounding the ascus.

Habit :—Goat dung.

14. *Ascobolus viridis.* (Curr.) Sacc. Syll. Fung. Vol. VIII, p. 519.

Apothecia gregarious, very variable in size, 2 to 3 mm. across, at first close, cup-shaped, becoming flat when mature, green, becoming yellowish green when mature, superficial, glabrous, leathery, at length ruptured at the apex by protruding asci; asci numerous arranged in vertical rows, cylindrical, lower portion distinguishable as stalk, 185

to 175·5 × 24·3 to 27 μ, octo-sporous, spores hyaline when young passing through all shades of violet to dark violet at maturity, oval, striate, ejected in a mass to some distance, 19 to 21·9 × 9·4 to 11 μ; paraphyses filiform, hyaline, surrounding the ascus, septate.

Habit :—Buffalo, Goat and Rabbit dung.

15. *Lasiobolus hirtellus.* (Karst.), Sacc. Syll. Fung., Vol. VIII, p. 538.

Apothecia gregarious, variable in size, superficial 279 μ in width (mean) 243 in height (mean); somewhat triangular, yellow, fleshy, setose seteae erect, arising from the sides, very few, 148 to 243 μ in height; asci few arranged in vertical rows, clavate, with a base, octosporous, 162 × 20 μ (mean); spores hyaline, smooth, uni-seriate-oval-globose, 13·5 to 16·2 × 10·8 μ.

Habit :—Goat dung.

16. *Lachnella fraxinicola.* (B. et Br.), Sacc. Syll. Fung., Vol. VIII, p. 396.

Apothecia scattered, sessile, scutellate, disc depressed, greenish yellow, covered with hairs, 311 to 337 μ in diam; hairs arising from the sides, pointed at the end and broad at the base, hyaline, unseptate, asci cylindrico-clavate, stalked, octo-sporous, 94·5 to 108 × 13·5 μ; spores fusoid, hyaline, in a single row, 13·5 to 18·5 × 6·4 μ; paraphyses filiform, hyaline, unseptate.

Habit :—Goat dung.

17. *Lachnella albido-fusca.* (Sacc.), Sacc. Syll. Fung., Vol. VIII, p. 397.

Apothecia scattered, sessile, globose, when young, becoming scutellate when mature, dark brown, covered with hairs; 135–202 μ across; hairs arising irregularly on the surface of the apothecium, dark yellow, unseptate; asci clavate; attenuated at the base, octo-sporous, 67·5 × 11 μ; spores ovato-globose, hyaline, arranged in a single row in the ascus, 6·2 to 7·8 × 6·2 μ; paraphyses simple unseptate, abundant, hyaline a little longer than the ascus.

Habit :—Goat dung.

18. *Coprinus niveus*, (pers), Fr. Carl. Ræ. Brit. Basid., p. 505. Pennington. Coprinus., p. 217.

Pileus 1·5 to 2 cm. across, elliptical, then campanulato-expanded, at length revolute, flesh white, very thin at the margin, almost persistently covered with snow-white floccose down; gills slightly attached, narrow, becoming blackish; stem 7 to 8 cm. high, 2 to 2·5 mm. thick, sub-equal or slightly attenuated upwards, villose, white, hollow; spores black, violet tinged, brownish when young, broadly elliptical, 13 to 16 × 10 to 12 μ, veil at the pileus is of mealy nature but tomentose at margin.

Habit :—Horse dung.

19. *Coprinus papillatus*, (Batsch.), Fr. Ræ, C. Brit. Basid. p. 507 (1922).

Pileus 5 to 10 mm. across, oval, then campanulate, finally, expanded, margins revolute, flesh dirty white, thin at the margin covered with hairs; gills slightly attached, narrow, becoming blackish at maturity; stem 1·9 to 3 cm. high, 8 to 10 mm. thick, smooth; lower portion root-like and brown upper dull white, hollow; spores black, brown when young, elliptic, 8 to 11 into 4·5 to 6·3 μ.

Habit :—Sambhar dung.

20. *Coprinus radiatus*, (Bolt.), Fr. Carlton. Rae. Brit. Basid., p. 512 (1922), Pennington. Coprinus p. 223.

Pileus 7 to 12 mm, across, at first ovate or short cylinderic, then campanulate, finally nearly or quite plane and slightly depressed at the center, very thin, deeply plicate, pileus with a few granular scales, slightly pruinose with glandtipped hairs, pale-brown or yellowish-brown; disc darker with bright yellow at the centre; gills narrow, distant, free; stem 2 to 2·8 cm. high, 1 to 1·5 mm. thick, slender, fragile, hollow, white becoming darker with age, slightly pruinose with glandular hairs; spores elliptical to broadly ovate, 8 to 9 × 6·5 to 7·5 mm., very dark.

Habit :—Horse and Goat dung.

21. *Coprinus ephemerus*, (Fr.), Carl. Rae. Brit. Basid., p. 515. 1922. Penn. Coprinus., p. 224.

Pileus 6 to 18 mm. across, ovate, then campanulate, finally expanded, often splitting and revolute, margin uneven sometimes, striate, plicate when expanded, very thin; disc even or slightly elevated, yellowish-brown, at first slightly pruinose with minute hairs, pileus smaller in dimensions than the bulb-like stem when young; gills linear, slightly adnexed or barely reaching the stem, usually white at the margin; stem 3 to 6 cm. high, 1 to 1·5 mm. thick, equal or slightly tapering upwards, hollow, wide; pores black in mass, ovate, cylindrico-elliptical, 8 to 10 × 6 to 8 μ.

Habit :—Rabbit dung.

22. *Bolbitus vitellinus*. (Pers.), Fr. Carl. Rae. Brit. Basid., p. 497, (1922).

Pileus 1·5 to 3 cm. across, yellowish-brown, submembranaceous, deeply campanulate, then expanding and convex, viscid, smooth, then furrowed, and splitting at the margin; stem 6·5 to 9·5 cm. high, 1.5 to 2·5 mm. thick, attenuated upwards from the sub-bulbous base, covered with white mealy fugacious flocci; gills ochraceous, then dark brown, free, attenuated at both ends, thin crowded; flesh yellowish-brown, thick at the disc; spores ferruginous, yellow under the microscope, broadly elliptical, 12·5 to 14 × 6·2 to 9 μ.

Habit :—Horse dung.

23. *Dendrostilbella byssina*. (Alb. et Schw.), Lindau. Die. Mikro. Pilze.
2 A B T. 2 A U f l. p., 244, fig. 310 (1922).

Plants scattered or more or less gregarious; stem simple, 1·2 to
1·5 mm. in height, stem straight or slightly flexuose, black, continues
to form head by branching variously, head globose, rust-grey in colour,
spores at the tips of the branches, elliptic to ovoid, smooth, light grey,
3·1 to 4 × 1·5 to 2·5 μ.

Habit :—Rabbit dung.

24. *Siliciopodium sanguineum*. (Corda.), Sacc. Eng. Prant. Nat. Pfl.
Fam. p. 490, 491. Fig. 254, i, c. I ABT.

Conidiophore compact, cylindrical, pure white, 7 to 1·5 mm. long,
175 to 324 μ thick; head somewhat swollen, conidia attached at the
tips of the hypheae on the head ; conidia hyaline, oval to elliptic, 20 to
24·3 μ in length, 11 μ in width.

Habit :—Buffalo dung.

25. *Oedocephalum glomerulosum*. (Bull.), Sacc. Syll. Fung. IV, 47, (1886)·

Gregarious at first pure white then tinged with rose colour, or
more frequently pale salmon colour with a tinge of yellow; stem sub-
cylindrical or slightly attenuated upwards, 94·5 to 200 × 10·8 μ;
transverse septa variable in number, sometime absent, inflated head
globose, verruculose, about 41 to 48 μ in diam., conidia elliptic, smooth,
13·5 to 16 × 6·5 to 10·8 μ.

Habit :—Buffalo, Goat and Horse dung.

26. *Arthrobotrys superba*. (Corda), Sacc. Syll. Fung. IV, 181, (1886),

Snow-white, glistening, densely gregarious and effused or more or
less scattered; fertile hypheae erect, up to 1/4 to 3/4 mm. high, simple,
septate, smooth, 4 to 5 μ in diam., delicate, bearing in the upper part
1 to many superposed whorls of conidia ; conidia arising from minute
blunt teeth on slight swellings of the conidiophore, crowded, one
septate, obovate, or oblong, lower cells usually the smaller, terminating
below into a minute point, 16 to 20 × 6·7 to 10.8 μ, smooth, hyaline.

Habit :—Rabbit dung.

27. *Stysanus stemonitis*. (Pers.), Corda, Sacc. Syll. Fung. IV, 621
(1886).

Stem scattered or more or less gregarious, simple, 1 to 1·3 mm.
high straight or slightly flexuose, head subcylindrical or lanceolate, at
first pale, becoming dark with age; conidia in long chains, ovoid to
elliptic, smooth, 3·1 to 3·6 × 2 5 μ.

Habit :—Rabbit and Sheep dung.

28. *Torula convoluta*. (Pers.), Lindau, Die. Mikro. Pilze. p. 202.
Fig. 231, p. 192.

Fertile, hyphae prostrata, pure white spreading, branched, branches bearing conidia in heads; heads globose; conidia compact, 27 μ across; conidia irregularly globose, minute, 6·2 μ in diam. (mean).

Habit :—Rabbit dung.

29. *Isaria brachiata*. (Batsch.), Schum., Engl. A. Prant., K. Teil, 2. I ABT., p. 490, 491.

Coremium cylindrical, erect, gregarious, white, simple, sometimes bi- tri- or tetra-fid near the apex, one to 1·2 cm. in height, ·5 to ·8 mm. in thickness. Conidia bearing hypheae arising from the sides, wooly ; conidia numerous, oval, hyaline, smooth, 3·1 to 3·7 μ in length, 1·5 to 2.5 μ in width.

Habit :—Sheep dung.

LAHORE,
9th September, 1932.

Explanation of Plates.   (Reduced to 1/3rd.)

PLATE I.

1. Pilobolus crystallinus (Wigg.).
    (*a*) sporangiophore ; (*b*) spores.
2. Pilobolus longipes (Van Tiegh.).
    (*a*) sporangiophore ; (*b*) spores.
3. Pilobolus minutus (Speg.).
    (*a*) sporangiophore ; (*b*) spores.
4. Magnusia nitida (Sacc.).
    (*a*) Perithecium ; (*b*) appendage ; (*c*) ascus.
5. Myxotrichum charatum (Kunz.).
    (*a*) glomerulus; (*b*) appendage ; (*c*) ascus.
6. Myxotrichum aeruginosum (Mont.)
    (*a*) glomerulus ; (*b*) appendage ; (*c*) ascus.
7. Sordaria macrospora (Auersw.).
    (*a*) perithecia ; (*b*) ascus.
8. Sordaria curvula (de Bary).
    (*a*) perithecia ; (*b*) ascus ; (*c*) spores.

PLATE II.

9. Chaetomium spirale (Zopf.).
    (*a*) perithecium ; (*b*) appendage ; (*c*) spores.
10. Ascobolus viridis (Curr.).
    (*a*) apothecia ; (*b*) ascus and paraphysis.

11. Sporomiella nigropurpurea (Ell. & Everh.)
      (*a*) perithecia; (*b*) ascus.
12. Pezolepis (Syd.) Sp.
      (*a*) apothecia; (*b*) ascus ; (*c*) paraphysis.
13. Lasiobolus hirtellus (Karst).
      (*a*) apothecium ; (*b*) ascus.
14. Coprinus ephemerus (Fr.).
15. Coprinus niveus (Fr.).
      (*a*) whole plant ; (*b*) basidium with spores.
16. Coprinus pappilatus (Batsch), Fr.
      (*a*) whole plant ; (*b*) basidium with spores.
17. Coprinus radiatus (Fr.).
      (*a*) whole plant ; (*b*) spores.
18. Bolbitus vitellinus. (Pers).
      (*a*) whole plant ; (*b*) spores.
19. Arthrobotrys superba (Corda).
20. Oedocephalum glomerulosum (Bull.).
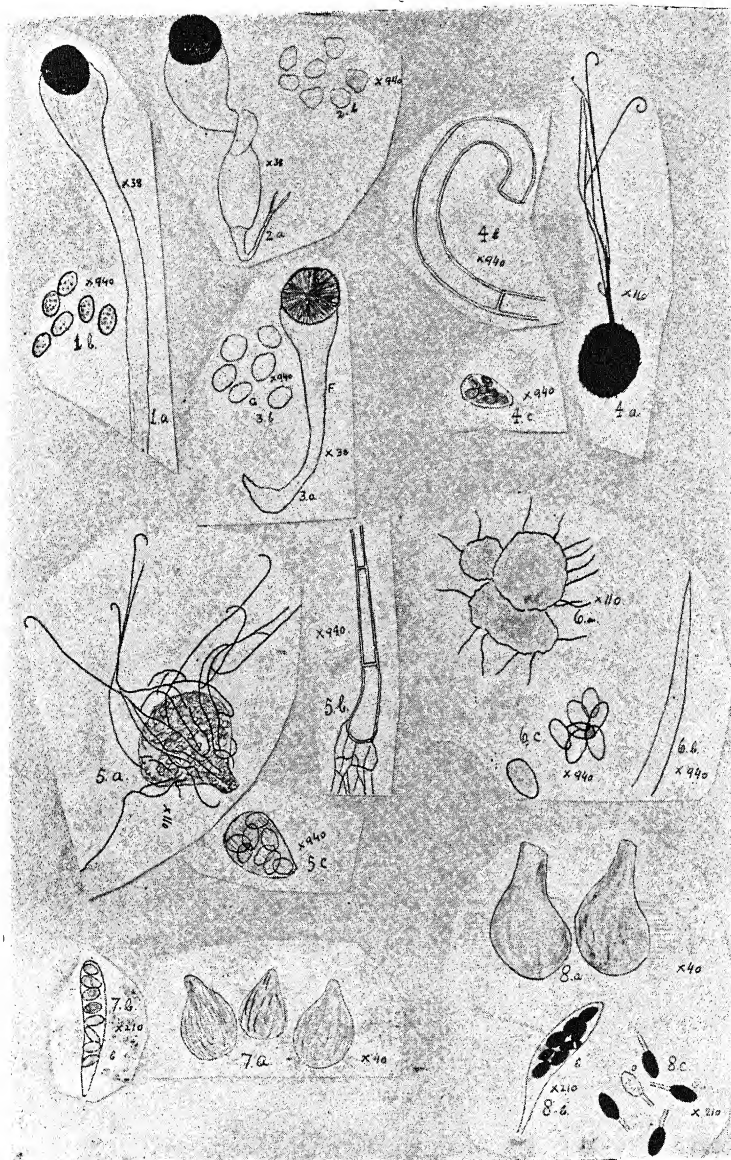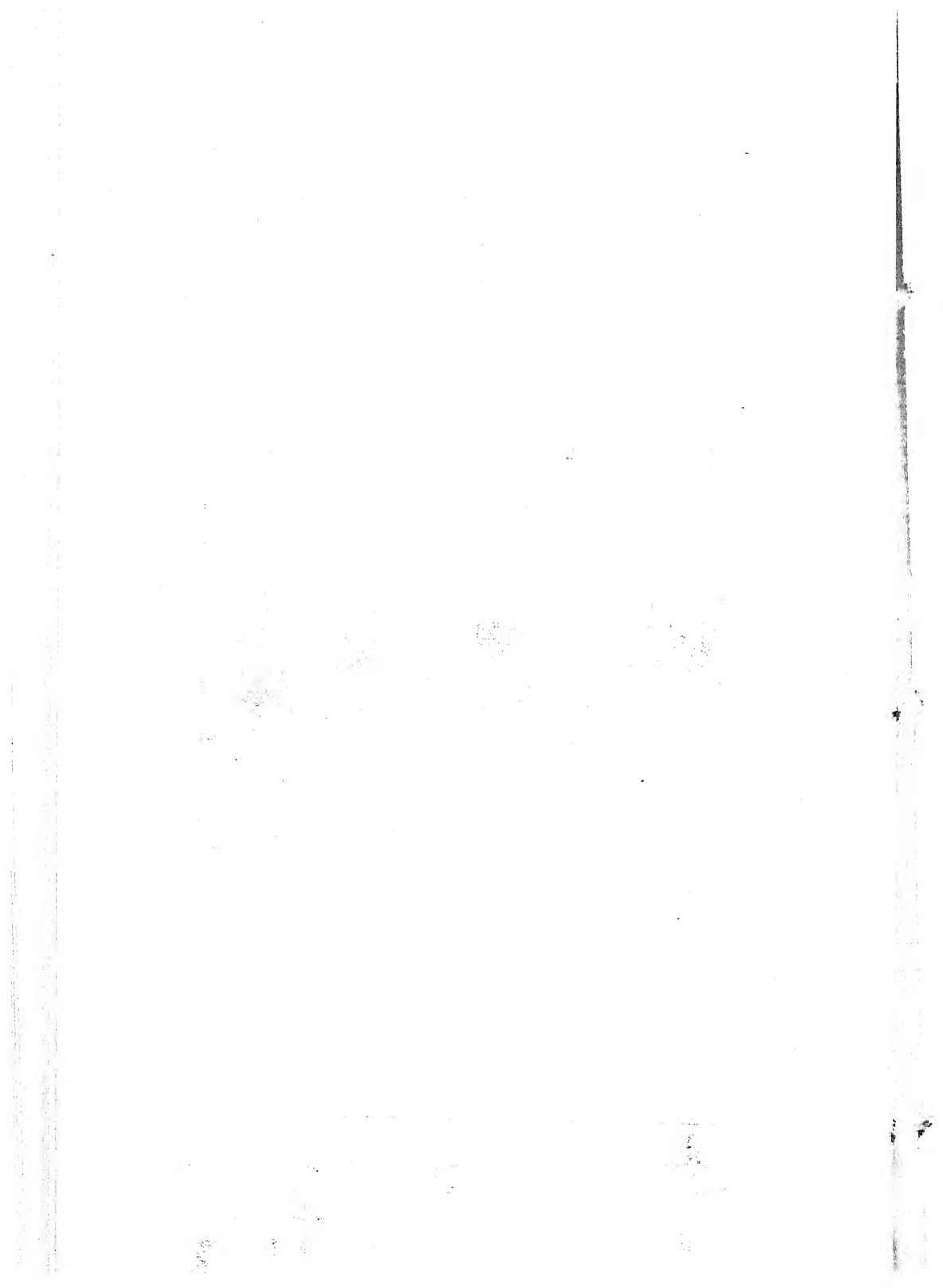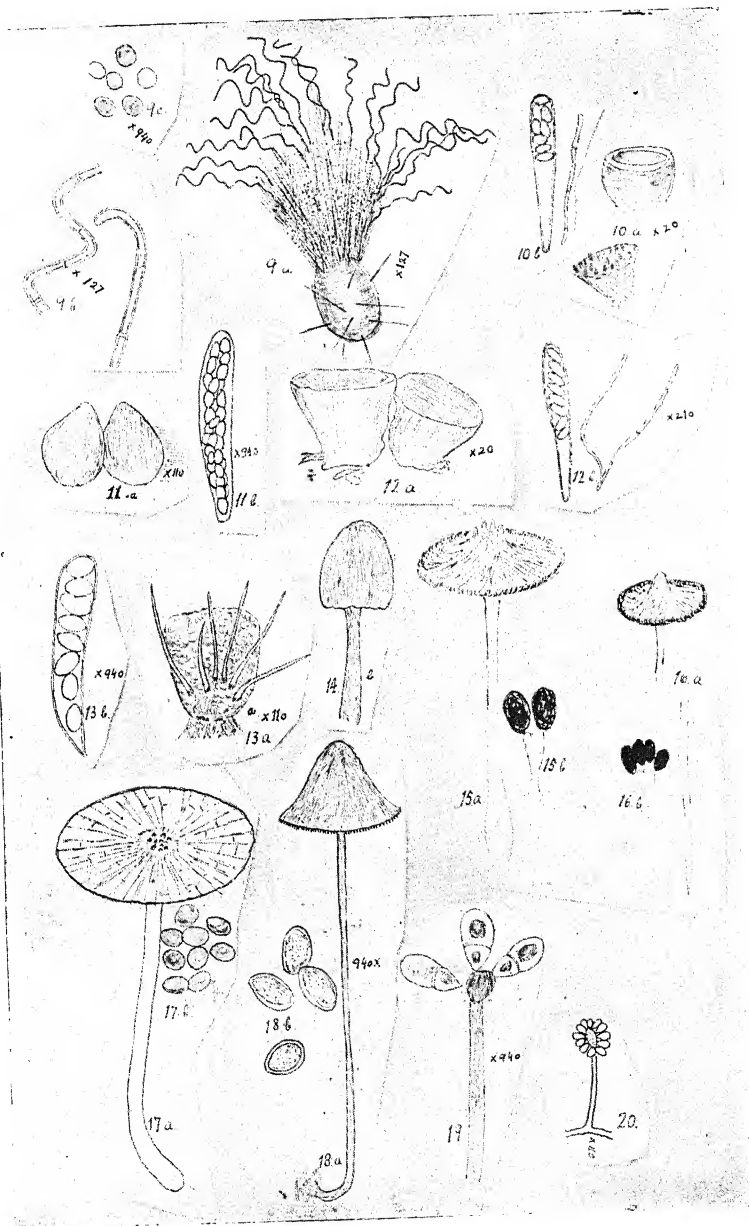
OBSERVATIONS ON THE BIOLOGY AND PHYSIOLOGICAL ANATOMY OF SOME INDIAN HALOPHYTES

BY

D. P. MULLAN,

St. Xavier's College, Bombay.

PART II.

Psammophilous Halophytes

Most of the plants described in this part are inhabitants of the sandy shore. As Schimper (8) remarks, the chief difficulties against which the psammophilous halophytes have to contend are: in fixing to a loose substratum; in obtaining water; and against the force of the sea wind. The plants of the sandy shore are annuals or perennials with mostly creeping or stunted stems and long roots. Some of the plants also occur inland as mesophytes, but the sand forms differ considerably in their configuration and structure from the inland forms.

Corchorus acutangulus, Lam. (Tiliaceae).

Among the sand plants the *Tiliaceae* has a representative in *Corchorus acutangulus*, Lam. (*C. aestuans*. Linn.). The plant is found growing both on the sands of the sea-shore and farther inland. The halophytic form (Fig. 104) shows a typical instance of dwarfing of the aerial parts; while the inland form (Fig. 105) shows luxuriant growth.

In the stem of the halophytic form, the cuticle is well developed and is coated with wax. The outer surface of the stem is covered with thick-walled, unicellular trichomes, each being inserted in a socket formed by the epidermal cells which, as a rule, hold tannin (Fig. 106). The outer cortex acts as the photosynthetic tissue. Large mucilage cavities are present in the cortex and the pith, being more numerous in the young parts (Fig. 107). A characteristic feature of the stele is the dilatation of some of the medullary rays at the cambial boundary (Fig. 108). The dilated part is composed of tangentially-elongated, thin-walled cells. As a rule, tannin is present in the outer cells making them appear like

a dark band on the top of each dilated part (Fig. 109). Except for the presence of a few starch grains, the cells of the dilated part are clear. The phloem appears stratified owing to the occurrence of secondary bast fibres. Tannin and aggregate crystals of oxalate of lime occur in: the epidermis, the cortex and the pith. The latter, in the old parts, is full of starch. Cork arises superficially in the stem and root.

Clothing hairs, similar to those of the stem, occur on the petiole, being more numerous on the adaxial surface. Unicellular trichomes are scattered on both surfaces of the leaf, being mainly confined to the region of the veins. The lower surface bears glandular hairs (Fig. 110). Stomata occur on both surfaces, being 75 per sq. mm. on the upper, and about 270 on the lower. The apparent increase in the number of the stomata is due to the reduction of the leaf surface. Both in surface extent (Fig. 111) and in depth (Fig. 113) the cells of the upper epidermis are more extensively developed than those of the lower (Fig. 112) and evidently act as water-reservoirs (6). In a lamina 0.15 mm. thick, the upper epidermis occupies 0.04 mm. The cuticle is thickly developed on both surfaces. The leaf is bifacial. Oil occurs in the mesophyll, being mainly confined to the palisade cells. Each minor vein is surrounded by a sheath of large, clear cells which run from epidermis to epidermis (Fig. 113). The uppermost cells of the sheath hold aggregate crystals, which thus occur in a row alongside the veins. Mucilage cavities occur in the petiole and in the region of the midrib and vary in number. (Fig. 114).

The anatomical structure of the plant growing inland shows some typical mesophytic characteristics. Thus in the stem, the chlorophyll tissue, the soft bast and the bast fibres are abundantly developed. The primary rays dilate only in the old parts and the cells hold starch. Tannin is mostly absent. The leaves of the mesophytic form are thin and show a well-developed chlorophyll tissue. The cells of the upper epidermis are less deep and the cuticle is less thickly developed (Fig. 115). A characteristic change, induced evidently by the mesophytic conditions, is to be seen in the lower epidermis. In the halophytic form, the lower epidermis appears, in surface view, to be composed of polygonal cells with straight walls (Fig. 112). Under mesophytic conditions, the cells are enlarged in surface extent and have irregularly wavy walls (Fig. 116). The trichomes are fewer in number. In the root, the secondary xylem is more porous.

Spermacoce hispida, Linn. (Rubiaceae).

Spermacoce hispida, Linn. is an annual herb growing both on the sea-shore and farther inland. The maritime forms are typically stunted in appearance, unbranched and have small leaves which are crowded together; while the inland forms are more luxuriantly developed.

In the stem of the halophytic form, the cuticle is well developed; its outer surface appears rough being coated with minute wart-like or stellate bodies (Fig. 117). In the young parts, trichomes occur all over the stem, but in the older parts, they are confined to the four angles of the quadrangular stem (Fig. 120). Stomata, with thick outer cuticular ridges, occur on the stem and are accompanied by subsidiary cells placed parallel to the pore (Fig. 117). The outer cortical cells act as the photosynthetic tissue. Oxalate of lime, in the form of raphides, aggregate crystals or irregular pieces, occur in the cortex and the pith. Tannin sacs are scattered in: the cortex, the secondary bast and the pith. At the base of each of the four ridges there occurs a clear cavity (*c*) which is surrounded by cells holding tannin. (Fig. 120). On the inner face of the xylem, at two opposite points, groups of pith cells get sparsely pitted and feebly lignified. The basal part of the stem is full of starch which occurs in the cortex, the medullary rays and especially in the pith.

In the leaf, the cuticle is well developed on the upper surface. Thick-walled trichomes cover both surfaces, being unicellular on the upper and uniseriate on the lower. Stomata occur on both surfaces, being 75 per sq. mm. on the upper (Fig. 121) and 125 on the lower (Fig. 122). The guard cells are accompanied by subsidiary cells, placed parallel to the pore, and have prominent outer cuticular ridges. The upper epidermis is composed of deep cells and seems to act as the aqueous tissue of the leaf (6). The lamina is 0.30 mm. thick, the upper epidermis being 0.06 mm. The leaf is bifacial (Fig. 123). Tannin and oil occur in the mesophyll cells. Idioblasts, holding raphides, are insinuated among the palisade cells. Crystal cells, each holding an aggregate crystal of oxalate of lime, occur below the first layer of palisade cells and run in a row parallel to the leaf surface. The smaller veins are ensheathed in a layer of clear cells. The epidermal cell, lying in the groove on the upper surface of the midrib, gets much enlarged and adds to the water-storing capacity of the upper epidermis. At the margin of the leaf the epidermal cells dilate considerably and form thick-walled marginal teeth with wide lumina (Fig. 124).

The anatomical structure of the mesophytic form differs in several respects. The ridges of the quadrangular stem are more prominently developed. Increased assimilatory activity is shown by the presence of chloroplasts in the cortex and by the fuller development of secondary soft bast. Tanniniferous cells are fewer in number. The upper epidermis of the leaf is less deep. A striking feature of the upper epidermis of plants living in shade is the peculiar thickening of the outer walls. In surface view, nearly all the cells are seen to bear at the centre a refractive body; in a cross section, these bodies are seen to be circumscribed areas of the outer walls which protrude in the form of plugs (Fig. 125). The latter are strongly developed in individuals living in shade, while in the halophytic forms they are absent. These bodies resemble the condensing portions of light-perceiving epidermal cells described by Haberlandt (3) who remarks: "it is more particularly those leaves which are constantly exposed to feeble illumination that stand in need of an upper epidermis with a well developed power of light perception." As previously noted, in *S. hispida*, it is only in individuals living in shade that the light-condensing apparatus is developed.

Launaea pinnatifida, Cass. (Compositae).

Launaea pinnatifida, Cass. is a perennial glabrous herb which thrives on the sandy sea-shore. In exposed situations by the sea, the plant shows the typical xerophytic characteristics; while in inland forms, these characteristics get mostly modified. In order to note the changes that may be induced under mesophytic conditions, specimens were taken from the sea-shore and planted in ordinary garden soil. After a period of nearly seven months, the specimens were removed for examination. Fig. 126 shows a typical halophytic form, while Fig. 127 shows the specimen grown under mesophytic conditions—both being photographed during the same dry season. The first peculiarity that strikes the eye is the great length of the roots in the halophytic form as compared with that of the mesophytic one. The former bears small thickish, glaucous leaves; while those of the inland form are fully-developed, thin and bright green.

In the halophytic form, the stem has a thick cuticle which is coated with wax. The epidermis and the outer cortical cells on the exposed side of the prostrate stem, hold anthocyanin. Stomata, with prominent outer cuticular ridges, occur on the stem and are sunk in pits. The latter are deeper on the exposed side of the

stem (Fig. 128) than on the side which is in contact with the soil (Fig. 129). Granules of wax overlie the guard cells upto the entry of the slit, thus plugging the outer opening. A comparison of Figs. 128 and 129 brings out the fact that the epidermal cells surrounding the stoma are deeper and the cuticle more thickly developed on the exposed side of the stem. The epidermis is followed by a 1–2 layered hypodermis which is composed of thick-walled, clear cells. The outer cortex is differentiated as the photo-synthetic tissue and consists of palisade parenchyma (Fig. 130). The palisade-like disposition of the cortical cells is confined mainly to the exposed side of the prostrate stem. Goebel (2) has shown that the shoot-axis whose leaves are few, or removed, has a palisade parenchyma much more developed; thus exhibiting an analogy with phylloclades. Sclerenchymatous elements, with wide lumina, occur in isolated groups in the cortex. In the old parts, the xylem masses get furrowed on their outer faces. In alcohol-preserved material all the xylem elements (the vessels as well as the paren-chyma) are seen to hold small inulin crystals, the latter being larger and more numerous at the nodes. Oil occurs in: the cortex, the medullary rays and the pith. The outer pith cells get lignified. Hanstein (5) and others have demonstrated an anomaly in the form of medullary phloem bundles and vascular bundles in the *Cichoriaceae*. In *L. pinnatifida* intraxylary phloem is present. Cork cambium arises subepidermally, in strips, on the exposed side, and leads to the formation of wound-cork. The structure and origin of the laticiferous tubes in the *Cichoriaceae* have been inves-tigated by de Bary (1), Van Tieghem (10) and others. In *L. pinnatifida*, the laticiferous tubes are confined to the outermost layer of the pericycle. In a cross section, the tubes occur in iso-lated groups forming arcs around the soft bast of each bundle. These constitute the main tubes, while similar, but smaller, elements are also scattered in the bast portion of the bundle. In a longi-tudinal section, the main tubes are seen to run parallel to the endodermis, giving out branches at intervals, but only towards the soft bast. From this it appears that the smaller tubes which, in a cross section, are seen to occur in the soft bast are the lateral branches of the main vessels. According to Haberlandt (3) and others, laticiferous tubes are conducting channels, while Warming (11) suggests that since they are mostly present in plants of hot, dry countries, one of their functions may be the protection of the plants against desiccation.

In the leaf, the cuticle is well developed and the outer surface is coated with wax. Tannin and oil occur especially in the upper epidermis. Stomata are equally abundant on both surfaces (Figs. 131, 132), being 125–150 per sq. mm. They are sunk in pits, the depression being more pronounced on the upper (Fig. 133) than on the lower surface (Fig. 134). The upper epidermis consists of deep cells (being 0.03 mm. in a leaf 0.45 mm. thick) and seems to act as the aqueous tissue (6). The leaf is bifacial. Starch is meagrely developed and oil occurs in the mesophyll cells. Laticiferous tubes follow the same course in the midrib and in the minor veins of the leaf as in the stem. In surface view, the major tubes are seen to branch out in all directions, the branches following closely the minor veins. After the monsoon, the old leaves turn yellow and become fleshy, attaining a thickness more than twice that of the functional leaves, and act as water-reservoirs (6).

The characteristic laticiferous tubes are also present in the root and occur in isolated groups among the sieve tubes. The medullary rays are prominently developed. The endodermis is composed of large cells with suberised walls. Crystal sand and oil occur in the cortex of the young parts. During the resting season especially, the tissues of the root get full of inulin which is seen to occur even in the xylem elements. In alcohol-preserved material the vessels are seen to be completely filled up with inulin crystals (Fig. 135). Owing to secondary growth there is a great development in the bast region, as a result of which the sieve tubes, with the laticiferous vessels, appear to be arranged more or less radially, being separated by the dilated parts of the medullary rays and the conjunctive tissue. Many of the sphaerites are seen to be deposited in the cells which are arranged around the smaller groups of laticiferous tubes (Fig. 136). In the tap root, very little inulin is present in the xylem elements, but it appears abundantly in the conjunctive tissue (Fig. 137). Cork arises in the pericycle. The tap root seems to act mainly as the storage organ for inulin and attains a diameter of 5–6 mm. The presence of inulin seems to explain the meagre development of starch in the leaves.

In the mesophytic form, the outer cortical cells do not assume the palisade-like form on the exposed side of the stem (Fig. 138). The chloroplasts are large and are arranged around the walls of the cortical cells. The respiratory cavities are wide. Oil and stone cells are rare and the conducting tissue is more fully developed. The cuticle of the leaf is feebly developed and the waxy

coating is lacking. The guard cells hold large chloroplasts with included starch. The lower epidermis shows the typical mesophytic modification and consists of comparatively large cells with wavy lateral walls (Fig. 139). The mesophyll cells are full of large chloroplasts with included starch; while oil and tannin are meagrely developed (Fig. 140). The spongy tissue is typical of a mesophyte, having wide intercellular spaces (Fig. 141). The lamina is thin, being half as thick as that of the halophytic form. The old leaves do not become thick and succulent (6). The roots remain thin, the tap root attaining a diameter of about 1.4 mm. Conjunctive tissue is feebly developed and inulin is not stored up under mesophytic conditions.

Scaevola Lobelia, Murr. (Goodeniaceae).

Scaevola Lobelia, Murr. (*S. Plumieri,* Vahl.) is a small, shrubby, glabrous plant with decumbent branches which root at the nodes. The plant spreads slowly along the sands giving rise to a sand-binding formation (Fig. 142). The thick, fleshy leaves arise close together at the end of the branches and assume a profile lie (Fig. 143).

In the stem, the cuticle is thickly developed. The outer surface of the epidermis, as well as the lateral walls, are coated with peculiar rod-shaped bodies of a waxy nature. The stomata are depressed and the outer cuticular ridges are thick and closely approximated (Fig. 144). The hypodermis acts as the photosynthetic tissue. At times, the intercellular spaces of the cortex hold mucilage. The cortical cells are feebly pitted and hold a few chloroplasts and oil. Towards the base of the stem, the cortical cells get tangentially stretched and show secondary division walls (Fig. 145). In the old parts, the pith gets pitted and lignified and the cells hold starch and oil. Aggregate crystals of oxalate of lime occur in the cortex and the pith. Cork arises subepidermally and leads to the formation of lenticels at the basal part of the stem.

The petiole repeats the general structure of the stem. At each side of the petiole, a mucilage cavity occurs in the cortex. The lamina is covered with wax. Solereder (9) has noted the presence of shortly-stalked, peltate glands. The number of the cells composing the shield of the gland varies from 4–8 (Fig. 146). The glands occur on both surfaces and are raised above the general leaf surface. They soon fall off and are mostly to be found on young leaves. The cuticle is thickly developed and the leaf shows

typical xerophytic characteristics. Stomata are equally abundant on both surfaces (Figs. 147, 148), being about 100 per sq. mm. They are depressed and the outer cuticular ridges are very thick and closely approximated (Fig. 149). The leaf structure is iso-lateral and consists of a hollow cylinder of palisade cells holding a prominently developed aqueous tissue. Starch is mostly absent in the leaf. The lamina is 1.8 mm. thick, the aqueous tissue occu-pying 0.97 mm. The aqueous tissue is composed of large thin-walled parenchymatous cells which, in the mature leaves, get elongated in the same manner as the palisade cells (Fig. 150). Except for a few minute aggregate crystals, the cells are mostly clear. Groups of enlarged and pitted terminal tracheides occur among the cells of the aqueous tissue. During the monsoon, the old leaves get very thick and store water as reserve material (6).

In the root, the narrow pith gets pitted and lignified. The endodermis consists of clear cells with suberised walls. The corti-cal cells are feebly pitted and in the old parts, get tangentially stretched and show secondary division walls. Oil is present in the cortex and medullary rays; while inulin occurs especially in the outer cortical cells. Cork starts superficially but later, cuts deeper into the cortex.

Ipomoea pes-caprae, Sweet. (Convolvulaceae).

Ipomoea pes-caprae, Sweet. (*I. biloba*, Forsk.) is, according to Schimper (8), the most widely spread plant of the sandy sea-shore. In exposed situations, the leathery leaves present the mar-gin to the sky by bringing the two lobes together (Fig. 151); while in shady situations the lobes expand a little. Seedlings were col-lected from the sea-shore and cultivated in ordinary soil, under mesophytic conditions, for nine months. The mesophytic form (Fig. 152) is weak having a very slender stem and small thin leaves. The lobes of the lamina are fully expanded, presenting the upper surface to the sky. The roots of the maritime form are typical of sand plants and attain a great length even in the seed-ling stage (Fig. 153). The rapid development of the root not only helps the seedling in reaching water, but also fixes it firmly to the sands and prevents it from being washed away by the rising tides.

The herbaceous stem is covered by a thick cuticle with a coat-ing of wax. Stomata are more or less even with the surface and the outer cuticular ridges are thickly developed (Fig. 154). The stomata are accompanied by subsidiary cells placed parallel to the

pore. Tannin occurs in the epidermal and subsidiary cells. Depressed glandular hairs are present on the stem (7). The cortex consists of three zones, viz: the outermost assimilatory tissue, the collenchyma and the parenchyma respectively (Fig. 154). Anthocyanin occurs in the first layer of the chlorenchyma and is mainly confined to the exposed sides of the stem. Vertical rows of crystal cells, each holding an aggregate crystal of oxalate of lime, occur in the cortex. Hallier (4) has noted the presence of a whitish sap in the axes of all the higher *Convolvulaceae*. In *I. pes-caprae*, both the collenchymatous and parenchymatous zones of the cortex are traversed by vertical rows of thin-walled cells which hold the milky-white sap (Figs. 155, 156). Hallier (4) has also noted the occurrence of starch in the white sap of *I. purpurea*, Lam. In *I. pes-caprae*, the milky sap is seen to be made up of at least two substances: a clear ground substance, which seems to be mainly mucilage, and large globules of oil (Fig. 156). The endodermis, in the young stem, holds chloroplasts. Islands of intraxylary phloem occur in the stem, each group being supported on its inner face by bast fibres. The first annual ring is normal but the second ring is not uniformly laid down. At first, only arcs of the second annual ring are formed, giving the xylem a lobed appearance. The xylem of the second ring is characterised by having wood parenchyma with wide luminae, the maximum diameter being 0.2 mm. In the old parts, these wide elements are filled up with thyloses, holding prominent nuclei and a few starch grains (Fig. 157). The medullary rays hold chloroplasts and starch. Secretory cells, holding the white sap, occur in the pith (Fig. 158). In the mature parts, the pith is fully loaded with large compound starch grains. Vertical rows of crystal cells, each holding an aggregate crystal of oxalate of lime, occur in the cortex and the pith. Cork develops subepidermally and lenticels are abundant on the basal part of the stem. A peculiarity of the basal part is that it is able to float in water, owing to the presence of lysigenously-formed air spaces.

In the petiole, the stomata are confined to the sides and secretory cells occur in the cortex. In the lamina, the lower epidermis is composed of large and deep cells. The cuticle is well developed and is coated with wax, both being more prominently developed on the lower surface which, owing to the approximation of the lobes, is the more exposed surface. Tannin occurs in the epidermis and is especially located in the cells of the margin. Depressed glandular hairs are to be found on both surfaces of the leaf (7),

Stomata occur on both surfaces (Figs. 159, 160), being 100 per sq. mm. on the upper and 75 on the lower surface. The occurrence of more stomata on the upper surface seems to be correlated with the fact that the leaf, by its position, exposes the lower surface and brings the upper into comparative shade. Subsidiary cells are placed parallel to the pore. The stomata are more or less even with the surface and the outer cuticular ridges are thickly developed (Fig. 161). The leaf structure is isolateral. The palisade tissue forms a hollow cylinder which is filled up by a clear, medullary, aqueous tissue. The latter is composed of large, thin-walled cells, most of them being elongated in the direction of the veins. The leaf is thicker towards the margin. In a lamina 0.6 mm. thick, the aqueous tissue occupies 0.3 mm. (6). Secretory cells, similar to those of the stem, occur in the aqueous tissue. Towards the lower surface (Fig. 162) the palisade cells are, as a rule, shorter and broader than those of the upper surface (Fig. 161). Rows of crystal cells occur in the neighbourhood of the veins and some also occur among the lower palisade cells. During the monsoon, the leaves are more fully developed and the lamina gets 1.1 mm. thick, the thickness being mainly due to the dilatation of the aqueous tissue (6).

In the root, secretory elements, similar to those of the stem, occur in the primary and secondary phloem and in the conjunctive tissue. A prominent feature of the inner cortex is the development of large air spaces separated by pluriseriate bridges. The spaces are lysigenous in origin and arise, as a rule, opposite each of the four primary phloem bundles. Thus the primary root of the seedling shows at first four air spaces. The primary root has a tendency to dilate at the distal end (Fig. 153); the dilatation is seen to be due to the expansion of the air spaces and to an increase in size and number of the cortical cells (Fig. 163). As the seedlings were found growing between tide marks, they must be getting submerged periodically. As is seen in Part I, the development of a lacunar cortex is a characteristic of the roots of plants which are submerged periodically. The medullary rays hold starch, oil and aggregate crystals. Cork arises at first superficially but later cuts deeper into the cortex so that in the old parts, the whole cortex gets peeled off, the root then acting mainly as a storage organ for starch. Lenticels arise at the proximal ends of old roots.

In the plant grown under mesophytic conditions, the stem is uniformly green, anthocyanin being absent. Tannin is not present in the epidermal cells of the leaves and the waxy coating is poorly

developed. The glandular hairs of the stem and leaf have meagre contents (7). Chlorenchyma is fully developed and the aggregate crystals are more numerous. In the leaf, the cells of the lower epidermis show somewhat wavy walls (Fig. 164). The palisade tissue on the upper side is typical (Fig. 165), but towards the lower surface, the cells are broad and loosely arranged (Fig. 166). The aqueous tissue is poorly developed, and the lamina is only 0.3–0.4 mm. thick (6). Air spaces are present in the root, showing, as in *A. ilicifolius,* that the lacunar cortex is a congenital structure. The secretory cells of the stem, leaf and root are present but are devoid of contents.

Neuracanthus sphaerostachys, Dalz, (Acanthaceae).

The *Acanthaceae* is represented among the psammophilous halophytes by *Neuracanthus sphaerostachys,* Dalz. The shoot of the typical halophytic form is stunted, the stems and leaves forming a compact cushion-like growth; while the plants living a short distance from the sea and in shade, show a more luxuriant growth (Fig. 167). After the monsoon, the aerial parts dry up, but the plant remains dormant and, in the next rainy season, sprouts up again. An examination of the plant during the dry season, reveals the presence of bud-like bodies at the surface of the soil. These are of the nature of hibernating organs and are very efficiently protected by dry hairy scales (Fig. 168). The buds remain dormant throughout the dry season and sprout on the advent of the monsoon.

The epidermis of the young stem is composed of thick-walled cells of various shapes, some of them holding oblique cystoliths (Fig. 169). The latter are sunk below the general outer surface. The stem is covered with uniseriate trichomes (Fig. 170). The epidermis is followed by a 3–4- layered hypodermis, interrupted by small groups of comparatively thin-walled cells forming a chlorenchyma (Fig. 171). The latter occur only in the young parts and are confined to the four corners of the obtusely quadrangular stem. Solerder (9) has noted the presence of inter- and intraxylary phloem. The pith is pitted and lignified. Acicular crystals occur in the pith and some of the outer cells hold cystoliths.

In the leaf, cystoliths occur on both surfaces and are disposed mainly around the veins. Each cystolith lies in a cell of a form similar to itself (Fig. 172). Glandular hairs (7) and uniseriate trichomes occur on the lower surface (Fig. 173). Stomata are

confined to the under surface of the leaf and are about 425 per sq. mm., while they are nearly half the number in the expanded leaves of the plants living in the shade—the apparent greater number in the former being due to the reduction of the leaf surface. The guard cells are accompanied by subsidiary cells placed transversely to the pore (Fig. 172). In a cross section, the upper epidermis forms a prominent feature, being composed of deep, clear cells which evidently act as the aqueous tissue (6). The lamina is 0.27 mm. thick, the upper epidermis being 0.06 mm. The leaf is bifacial and palisade cells hold oil (Fig. 174).

Long and persistent root hairs clothe the greater part of the root. The cortex is supported by short sclerenchymatous cells. An anomaly, in the form of intraxylary phloem, occurs among the secondary xylem. The pith is pitted and lignified.

The internal structure of the plants growing in shade do not show any marked change except that the chlorenchyma is more fully developed and starch is abundantly present. In the root, the acicular crystals and cystoliths are more numerous.

Lepidagathis trinervis, Nees. (Acanthaceae).

Lepidagathis trinervis, Nees. is another member of the *Acanthaceae* which grows on the sandy sea-shore. After fruiting, the aerial shoot dies down but the capsules remain fixed to the soil. Throughout the dry season, the fruits and seeds remain efficiently protected by the dry bracts and bracteoles. On the advent of the monsoon, the seeds germinate in the (now moist) bracts and produce new plants around the withered remains of the bracts of the last generation.

The prostrate branches are quadrangular, the angles being produced into wings (Fig. 175). The epidermal cells are vertically elongated and are deep and clear. Cystoliths occur in some cells and are, as a rule, more numerous on the shorter sides of the branch. The epidermis is followed by 3–4 layers of collenchyma which is succeeded by a parenchymatous cortex. Rod-shaped crystals of oxalate of lime occur in the cortex and the pith. The distal end of the wings are stiffened by collenchyma, while the proximal parts act as the photosynthetic tissue (Fig. 176). Thus the chlorenchyma is confined to the four corners of the quadrangular branches (Fig. 175). The stomata occur in the region of the chlorenchyma. The guard cells have very thick walls and the outer cuticular ridges are prominently developed (Fig. 177). Solereder (9) has noted the occurrence of inter- and intraxylary

phloem in: *L. cuspidata*, Nees., *L. prostrata*, Dalz. and *L. termi-nalis*, Hochst. Such an anomaly is also found in *L. trinervis*, Nees.

The leaves are small; the two halves of each lamina approxi-mate, thus presenting the margin to the sky. Depressed glandular hairs, resembling those of the previous plants, occur on both sur-faces of the leaf (Figs. 178, 179). Each gland (Fig. 180) consists of a short stalk-cell and a rounded head which is divided by vertical walls into 4–8 cells. The upper epidermis consists of deep, clear cells which seem to act as the main aqueous tissue. The lamina is 0.19 mm. thick, the upper epidermis occupying 0.04 mm. Stomata occur on both surfaces of the reduced leaves, being 175 per sq. mm. on the upper and 275 on the lower. The guard cells are accompanied by small subsidiary cells, placed parallel to the pore, and have thick walls with well-developed outer cuti-cular ridges. (Fig. 181). Spindle-shaped cystoliths lie in cells of forms similar to themselves, their greatest diameter running parallel to the surface of the leaf (Fig. 182). The leaf is bifacial and possesses strongly developed marginal teeth (Fig. 183).

Clerodendron inerme, Gaertn. (Verbenaceae).

Clerodendron inerme, Gaertn. is a straggling shrub which is often found along with the mangroves on the verge of high-water mark where its roots are washed by the rising tides. It thrives equally on wet or dry saline soil, as well as under mesophytic con-ditions. A plant was taken from the sea-shore and cultivated under mesophytic conditions for ten months. Figs. 184 and 185 are photographs (taken in the dry season) of the halophytic and mesophytic forms respectively. In the halophytic form, the small, thick leaves arrange themselves on one side of the branch, thus assuming a profile position. In the mesophytic form, the leaves are broad and thin and, standing wide apart, present their full upper surface to the light.

The young stem of the halophytic form has a thick cuticle which is coated with wax. Anthocyanin occurs in the epidermis of the exposed part. Oxalate of lime occurs in the epidermis and the forms of crystals are very diverse, being simple, aggregate, rod-shaped, etc. Unicellular and uniseriate trichomes (Fig. 186), as well as glandular hairs (Fig. 187), occur on the stem. The hypodermis consists of 1–2 layers of more or less collenchymatous cells with wide lumina. Except for a few starch grains, the hypodermis is clear and is supported by vertically elongated,

lignified cells. The latter occur singly in the young parts but, in the old parts, they form small isolated groups. Chloroplasts occur in the primary cortex. The primary medullary rays and the pith are composed of thick-walled, pitted and lignified cells, holding starch grains. The phellogen layer arises below the hypodermis (Fig. 188) and the cork is composed of lignified, phelloid cells. In the case of plants living on the verge of high-water mark, the lower part of the stem bears prominent lenticels and develops a lacunar secondary cortex which is supported by groups of stone cells.

In the petiole, the hypodermal and cortical cells towards the abaxial side are especially enlarged and seem to act as the aqueous tissue. The stomata and chlorenchyma are confined to the sides (Fig. 189). Simple crystals of oxalate of lime occur in the upper epidermis of the leaf. The cuticle is well developed and the outer surface is coated with wax. Depressed glandular hairs (Fig. 190) occur on both surfaces, being more numerous on the lower (7). Stomata are very rare on the upper epidermis (Fig. 191), while on the lower surface (Fig. 192) they are about 125 per sq. mm. The stomata are even with the surface and the outer cuticular ridges are well developed and closely approximated (Fig. 194). The upper epidermis is deep and seem to act as the aqueous tissue, being 0.03 mm. in a leaf 0.48 mm. thick (6). The leaf structure is bifacial (Figs. 193, 194). Oil occurs in the mesophyll, being more prominent in the spongy tissue. Trichomes, which are mostly absent on the leaf, are confined to the region of the main vein, being more numerous on the upper surface. At the end of the monsoon, the old leaves get thick and succulent, being nearly thrice as thick as the functional leaves. Such leaves are pale yellow in colour and act mainly as water-reservoirs. (6).

In the root, the primary cortex is lacunar and resembles somewhat that of the mangroves. In the case of individuals whose roots are washed by the tides, the outer cortical cells have a tendency to form lysigenous cavities. In the young roots, the inner cortical cells are in an active state of division. As growth proceeds, the cells become irregular in outline, with the consequence that the intercellular spaces increase in size. Thus the cortical lacunae are both of schizogenous and lysigenous origin (Fig. 195.). The main lacunae are wide and radially placed. In its lacunar cortex, *C. inerme* shows its affinity with the plants living in the salt swamp. As in *Acanthus ilicifolius*, the lacunar cortex seems to be a congenital structure, for it persists even in individuals

which live in well-drained soil. The medullary rays and the pith are pitted and lignified. The phellogen arises in the pericycle and cork is composed of more or less lignified, phelloid cells as in the mangroves.

In the mesophytic form, photosynthetic activity is more vigorous and the cortex is full of chloroplasts with included starch. The pith is not lignified throughout. In the leaf, the cuticle and the waxy coating are poorly developed and the glands are mostly devoid of contents (7). The lower epidermis, in surface view, is no longer composed of small cells with straight walls (Fig. 192), but consists of large cells with irregularly wavy walls (Fig. 196). The lamina is thinner than that of the halophytic form, being 0.27 mm. thick; and the old leaves do not become thick and succulent (6). The spongy tissue is very loosely built (Fig. 197) and oil is not abundantly developed.

Literature Cited.

1. BARY, A. DE—Comparative Anatomy of the vegetative organs of the Phanerogams and Ferns. Engl. Ed., Oxford, 1884, p. 524.

2. GOEBEL, K.—Organography of Plants. Engl. Ed., Oxford, 1905, pp. 281, 445.

3. HABERLANDT, G.—Physiological Plant Anatomy. Engl. Ed., 1928, pp. 328, 340-41, 623.

4. HALLIER.—Naturl. Glied d. c. Engler's Bot. Jahrb. xvi, 1893, pp. 453-591.

5. HANSTEIN, R.—Uber die Organe der Harz-und Schleimabsonderung in den Laubknospen. Bot. Zeitg., xxvi, 1868, pp. 734 *Sqq.*

6. MULLAN, D. P.—Observations on the water-storing devices in the leaves of some Indian Halophytes. Journ. Ind. Bot. Soc., x, 2, pp. 126-33.

7.   „   .—On the occurrence of glandular hairs (salt-glands) on the leaves of some Indian Halophytes Journ. Ind. Bot. Soc., x, 3, pp. 184-89.

8. SCHIMPER, A. F. W.— Plant Geography upon a physiological basis. Engl. Ed., Oxford, 1903, p. 181.

9. SOLEREDER, H.—Systematic Anatomy of the Dicotyledons. Engl. Ed., Oxford, 1908.

10. TIEGHEM, P. VAN.—Canaux secret. Ann. Sc. Nat., ser. 5, xvi, 1872 pp. 97-147.

11. WARMING, E.—Oecology of Plants. Engl. Ed., Oxford, 1909 p. 125.

Explanation of Plates.

The initial magnification is indicated after each figure. All the figures have been reduced to about one-third in reproduction.

Corchorus acutangulus, Lam.

Fig. 104.—Photograph, showing the halophytic form.

Fig. 105.—Photograph, showing the mesophytic form.

Fig. 106.—T. S. stem, showing a trichome. ($\times$ 360).

Fig. 107.—T. S. stem : m, mucilage cavity. ($\times$ 240).

Fig. 108.—Photomicrograph. T. S. stem. ($\times$ 82).

Fig. 109.—T. S. stem, showing a medullary ray. ($\times$ 240).

Fig. 110.—A glandular hair on the lower surface of the leaf. A, in surface view; B, in T. S. ($\times$ 500).

Fig. 111.—Leaf upper epidermis. ($\times$ 240).

Fig. 112.—Leaf : lower epidermis. ($\times$ 240).

Fig. 113.—T. S. leaf : u, upper epidermis; p, palisade cells holding oil. ($\times$ 500).

Fig. 114.—T. S. leaf, showing the region of the main vein : m, mucilage cavities. Semi-diagrammatic. ($\times$ 82).

Fig. 115.—T. S. leaf : mesophytic form. ($\times$ 500).

Fig. 116.—Leaf : lower epidermis. Mesophytic form. ($\times$ 240).

Spermacoce hispida, Linn.

Fig. 117.—Stem : epidermis. ($\times$ 360).

Fig. 118.—T. S. stem : a trichome. ($\times$ 240).

Fig. 119.—T. S. stem, showing the cortex. ($\times$ 360).

Fig. 120.—T. S. stem, showing a ridge. ($\times$ 240).

Fig. 121.—Leaf : upper epidermis. ($\times$ 240).

Fig. 122.—Leaf : lower epidermis. ($\times$ 240).

Fig. 123.—T. S. leaf : u, upper epidermis. ($\times$ 500).

Fig. 124.—T. S. leaf, showing the marginal teeth. ($\times$ 240).

Fig. 125.—T. S. leaf, mesophytic form : u, upper epidermis. ($\times$ 360).

Launaea pinnatifida, Cass.

Fig. 126.—Photograph : halophytic form.

Fig. 127.—Photograph : mesophytic form.

Fig. 128.—T. S. stem : exposed side. ($\times$ 500).

Fig. 129.—T. S. stem : side in contact with the soil. ($\times$ 500).

Fig. 130.—T. S. stem, showing the palisade-like cortical cells. ($\times$ 500).

Fig. 131.—Leaf : upper epidermis. ($\times$ 240).

Fig. 132.—Leaf : lower epidermis. ($\times$ 240).

Fig. 133.—T. S. leaf: *u*, upper epidermis ; *p*, palisade cells holding oil. (× 500).

Fig. 134.—T. S leaf, showing the spongy tissue. (× 500).

Fig. 135.—Photomicrograph. T. S. root, showing the xylem elements holding inulin crystals. (× 5C0).

Fig. 136.—T. S. root: conjunctive tissue showing a group of laticiferous tubes and inulin crystals. (× 500).

Fig. 137.—Photomicrograph. T. S. root, showing the conjunctive tissue holding masses of inulin crystals. (× 82).

Fig. 138.—T. S. stem: mesophytic form. (× 500).

Fig. 139.—Leaf: lower epidermis of mesophytic form. (× 240).

Fig. 140.—T. S. leaf, mesophytic form : *u*, upper epidermis. (× 240).

Fig. 141.—T. S. leaf, mesophytic form : *l*, lower epidermis. (× 240).

Scaevola Lobelia, Murr.

Fig. 142.—Photograph : a *S. Lobelia* formation.

Fig. 143.—Photograph : profile position of leaves.

Fig. 144.—T. S. stem. (× 500).

Fig. 145.—T. S. stem, showing the cortex. (× 240).

Fig. 146.—A glandular hair from the under surface of the leaf. (× 500).

Fig. 147.—Leaf : upper epidermis. (× 240).

Fig. 148.—Leaf lower epidermis. (× 240).

Fig. 149.—T. S. leaf: *u*, upper epidermis ; *p*, palisade cells. (× 500).

Fig. 150.—T. S. leaf : *p*, palisade cells ; *a*, aqueous tissue. (× 240).

Ipomosa pes-caprae, Sweet.

Fig. 151.—Photograph, showing the position of leaves ; halophytic form.

Fig. 152.—Photograph : mesophytic form.

Fig. 153.—Photograph : seedling.

Fig. 154.—T. S. stem, showing the cortex. (× 500).

Fig. 155.—T. S. stem, showing the inner cortex with a secretory cell, *s*. (× 240).

Fig. 156.—L. S. hypocotyl, showing the secretory cells (*s*) in the cortex : *o*, oil ; *g*, starch grains. (× 500).

Fig. 157.—T. S. stem : secondary xylem. (× 240).

Fig. 158.—T. S. stem : pith with a secretory cell, *s*. (× 240).

Fig. 159.—Leaf: upper epidermis. (× 240).

Fig. 160.—Leaf: lower epidermis. (× 240).

Fig. 161.—T. S. leaf : *u*, upper epidermis. (× 500).

Fig. 162.—T. S. leaf : *l*, lower epidermis. (× 500).

Fig. 163.—Photomicrograph. T. S. root of seedling. (× 82).

Fig. 164.—Leaf mesophytic form : lower epidermis. (× 240).

Fig. 165.—T. S. leaf, mesophytic form : u, upper epidermis. (× 500).

Fig. 166.—T. S. leaf, mesophytic form : l, lower epidermis. (× 500.)

Neuracanthus sphaerostachys, Dalz.

Fig. 167.—Photograph : A, halophytic forms : B, mesophytic form.

Fig. 168.—Photograph, showing the hibernating organs.

Fig. 169.—T. S. stem, showing the epidermis with cystoliths, K. (× 240).

Fig. 170.—T. S. stem, showing a trichome. (× 240).

Fig. 171.—T. S. young stem : h, hypodermis ; c, chlorenchyma. (× 240).

Fig. 172.—Leaf : lower epidermis ; k, cells holding cystoliths. (× 240).

Fig. 173.—Leaf : a trichome on a vein. (× 180).

Fig. 174.—T. S. leaf : u, upper epidermis ; p, palisade cells. (× 240).

Lepidagathis Trinervis, Nees.

Fig. 175.—T. S. stem. Semi-diagrammatic : c, chlorenchyma. (× 51).

Fig. 176.—T. S. stem (region of the wing) : c, chlorenchyma. (× 240).

Fig. 177.—T. S. stem, showing a stoma. (× 560).

Fig. 178.—Leaf : upper epidermis. (× 360).

Fig. 179.—Leaf : lower epidermis ; g, gland. (× 360).

Fig. 180.—T. S. leaf, showing a glandular hair. (× 500).

Fig. 181.—T. S. leaf : u, upper epidermis ; k, cystoliths. (× 500).

Fig. 182.—T. S. leaf, showing the upper epidermis with a cystolith. (× 180).

Fig. 183.—T. S. leaf, showing the marginal teeth. (× 240).

Clerodendron inerme, Gaertn.

Fig. 184.—Photograph, showing the profile positon of leaves in the halophytic form.

Fig. 185.—Photograph : mesophytic form.

Fig. 186.—T. S. stem : t, trichome ; s, stone cell. (× 500).

Fig. 187.—A glandular hair on the stem : A, in surface view ; B, in cross section. (× 500).

Fig. 188.—T. S. old stem, showing cork formation. (× 500)

Fig. 189.—T. S. petiole, showing the region of the chlorenchyma. (× 500).

Fig. 190.—T. S. leaf : a gland on the upper epidermis. (× 500).

Fig. 191.—Leaf : upper epidermis. (× 500).

Fig. 192.—Leaf : lower epidermis. (× 500).

Fig. 193.—T. S. leaf : u, upper epidermis ; p, palisade cells. (× 240).

Fig. 194.—T. S. leaf : l, lower epidermis ; s, spongy tissue. (× 500).

Fig. 195.—Photomicrograph. T. S. root, showing the lacunar cortex. (× 240).

Fig. 196.—Leaf : lower epidermis of the mesophytic form. (× 500).

Fig. 197.—T. S. leaf : mesophytic form. (× 240).

PLATE VI.

144

149

150

145

146

147

154

155

148
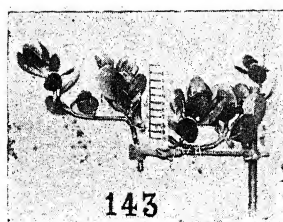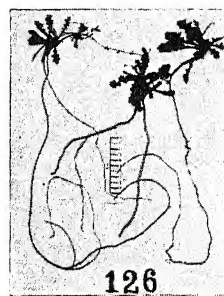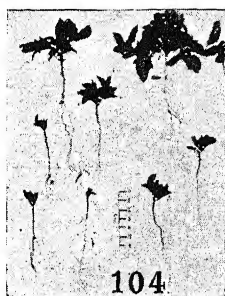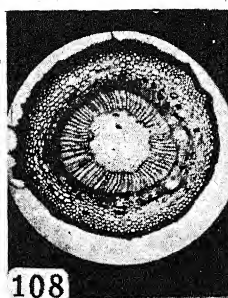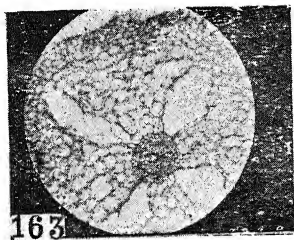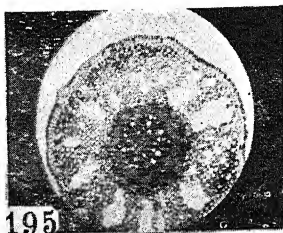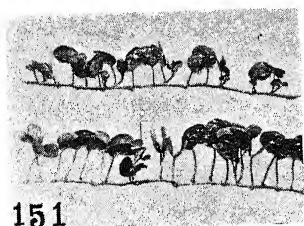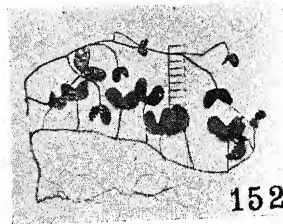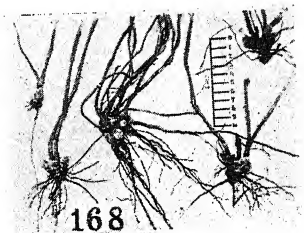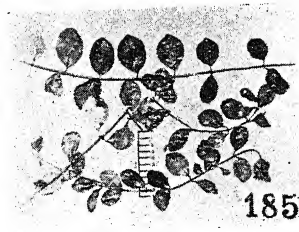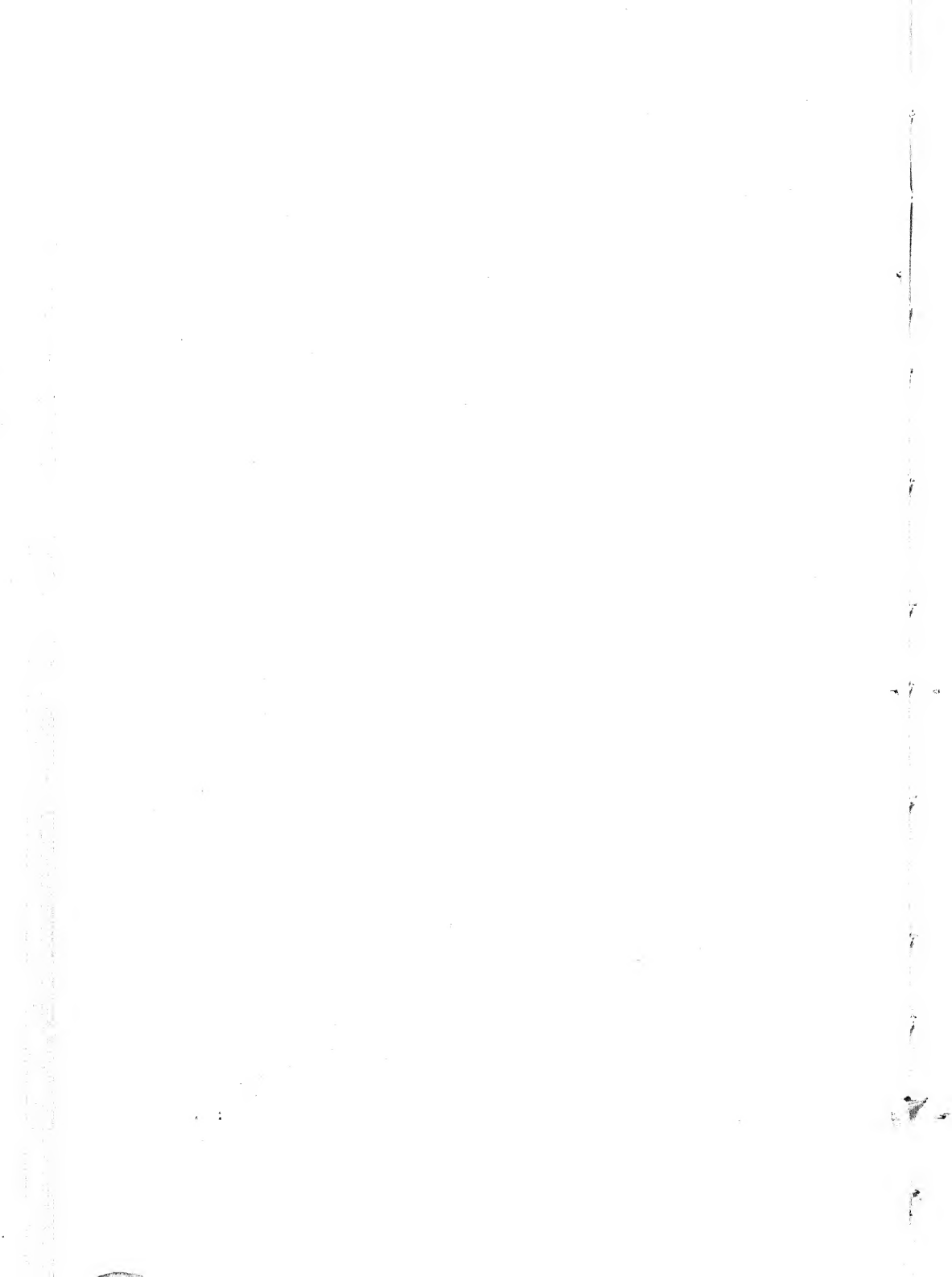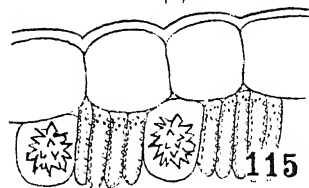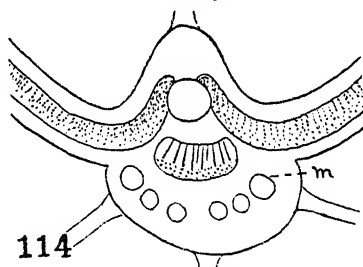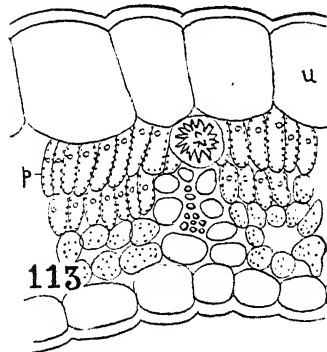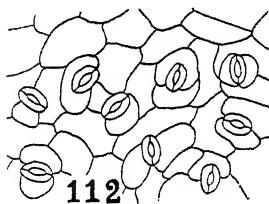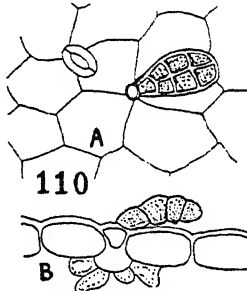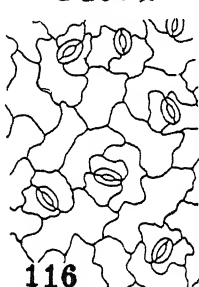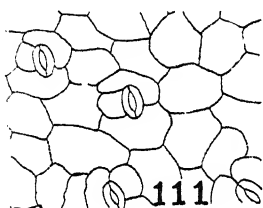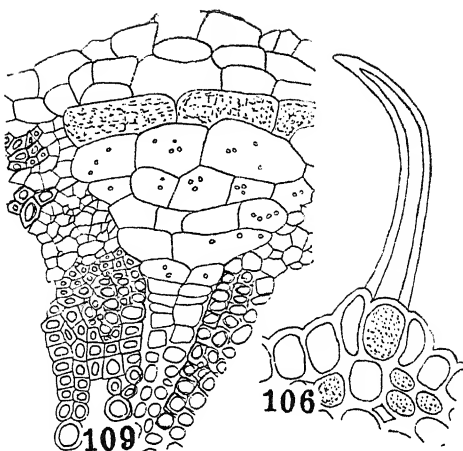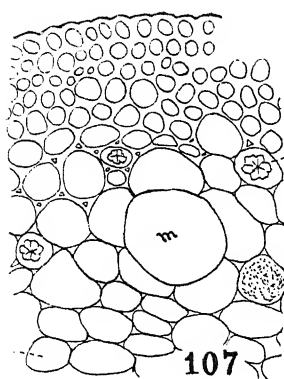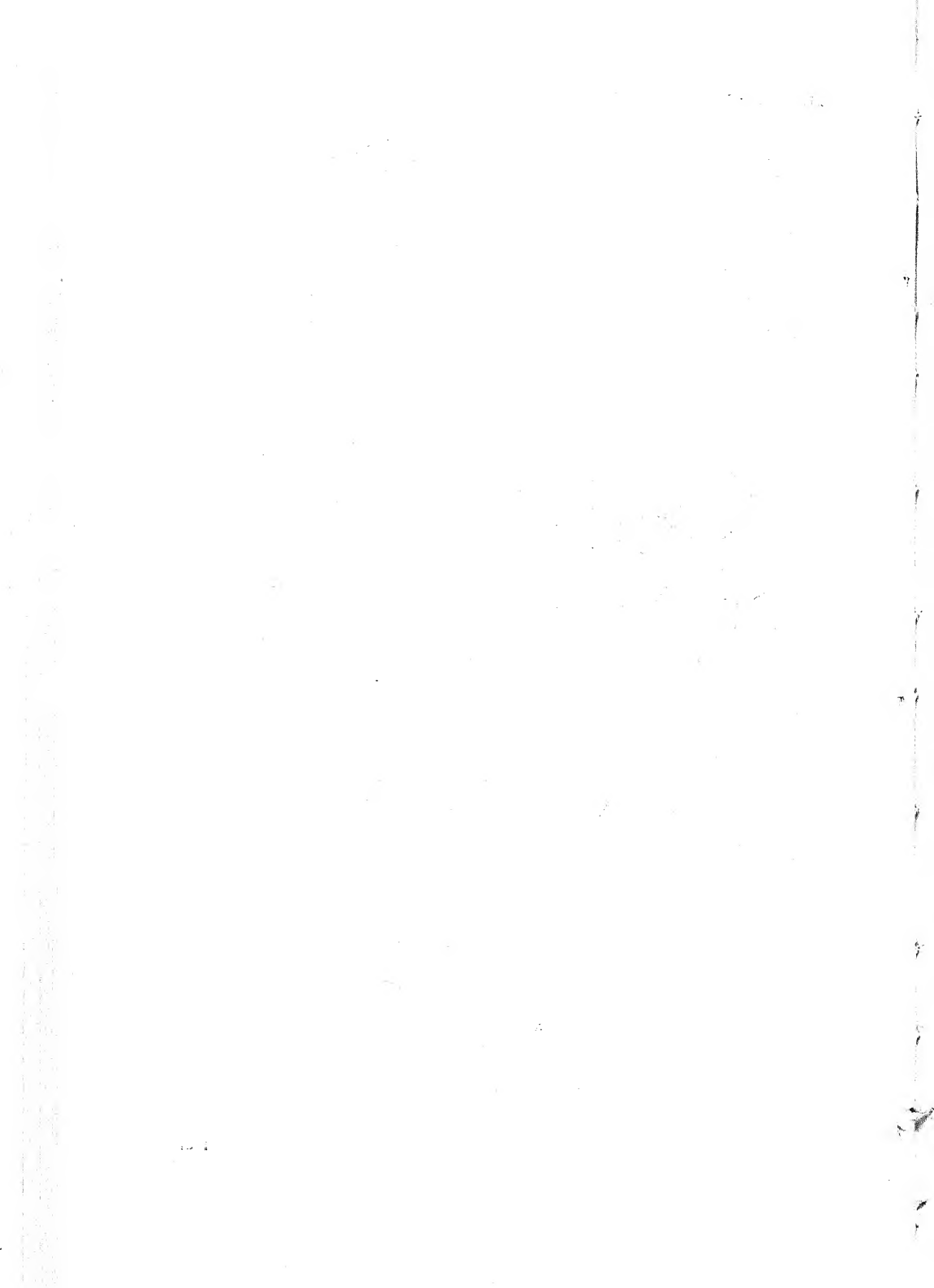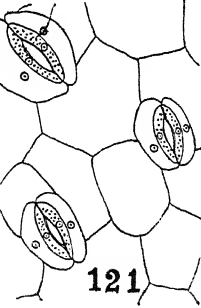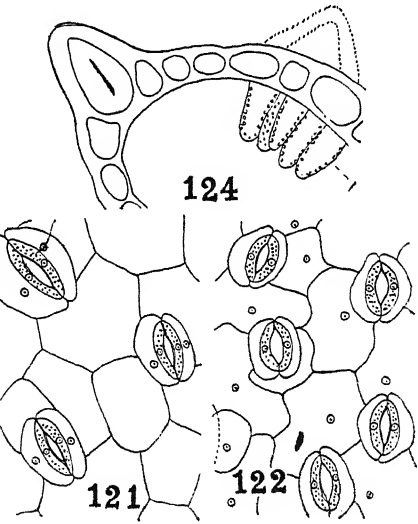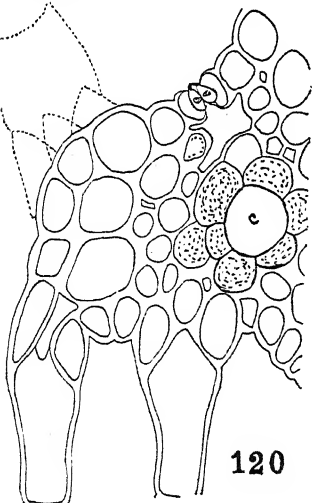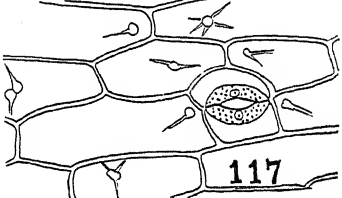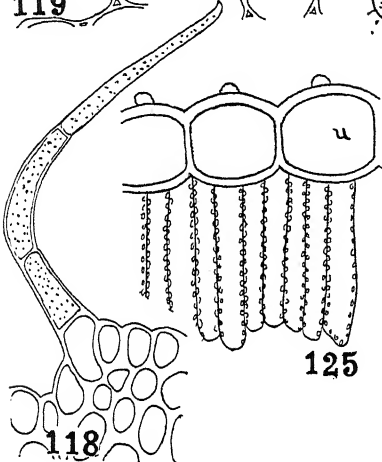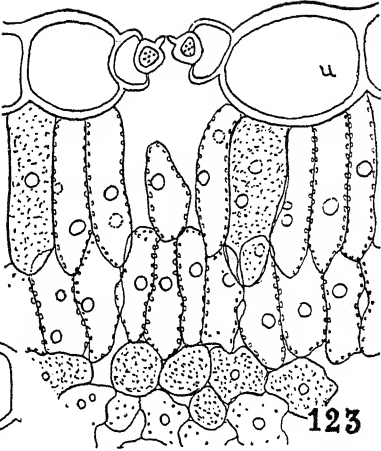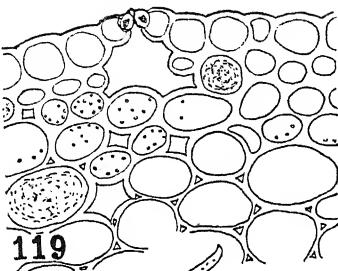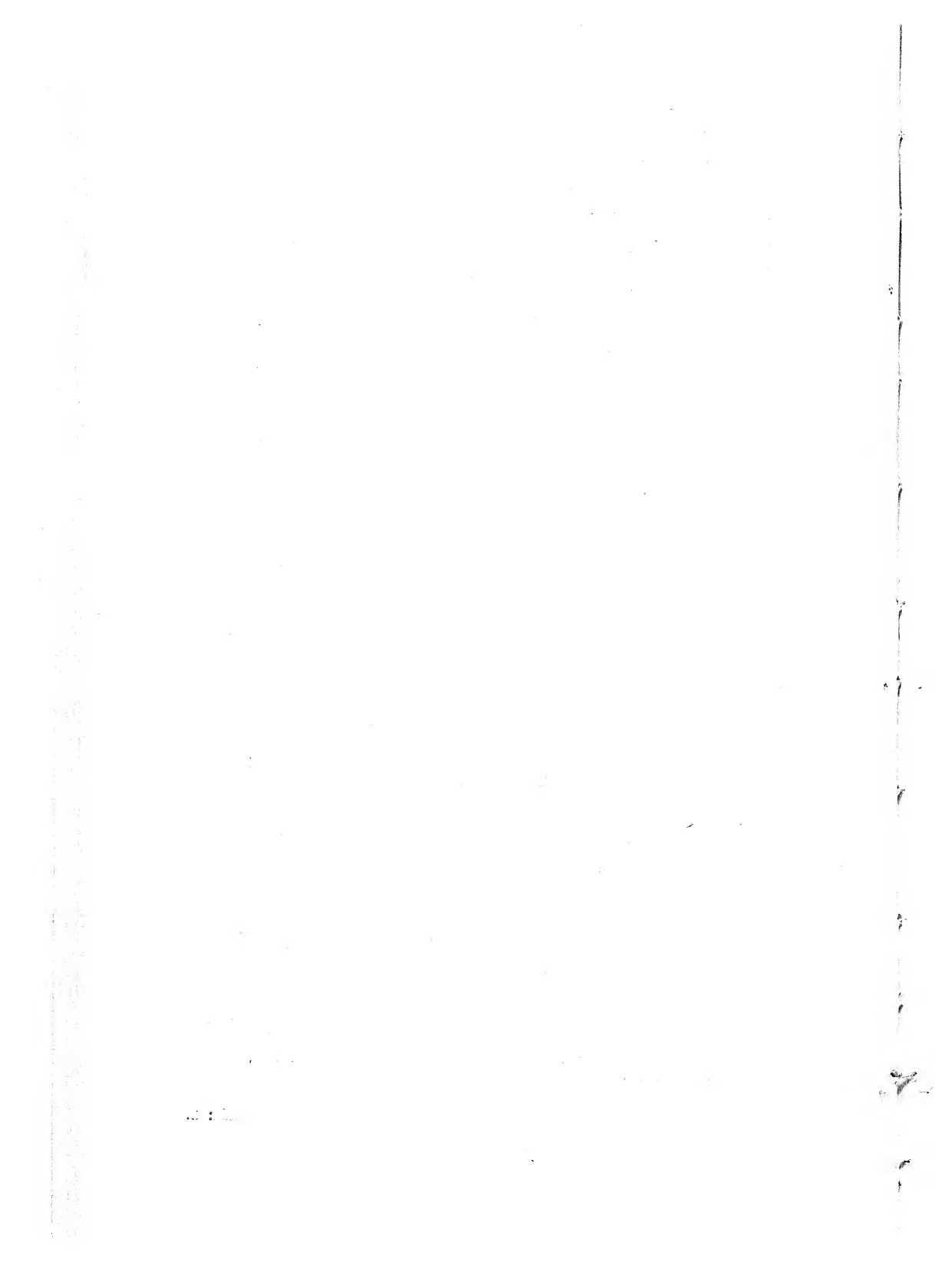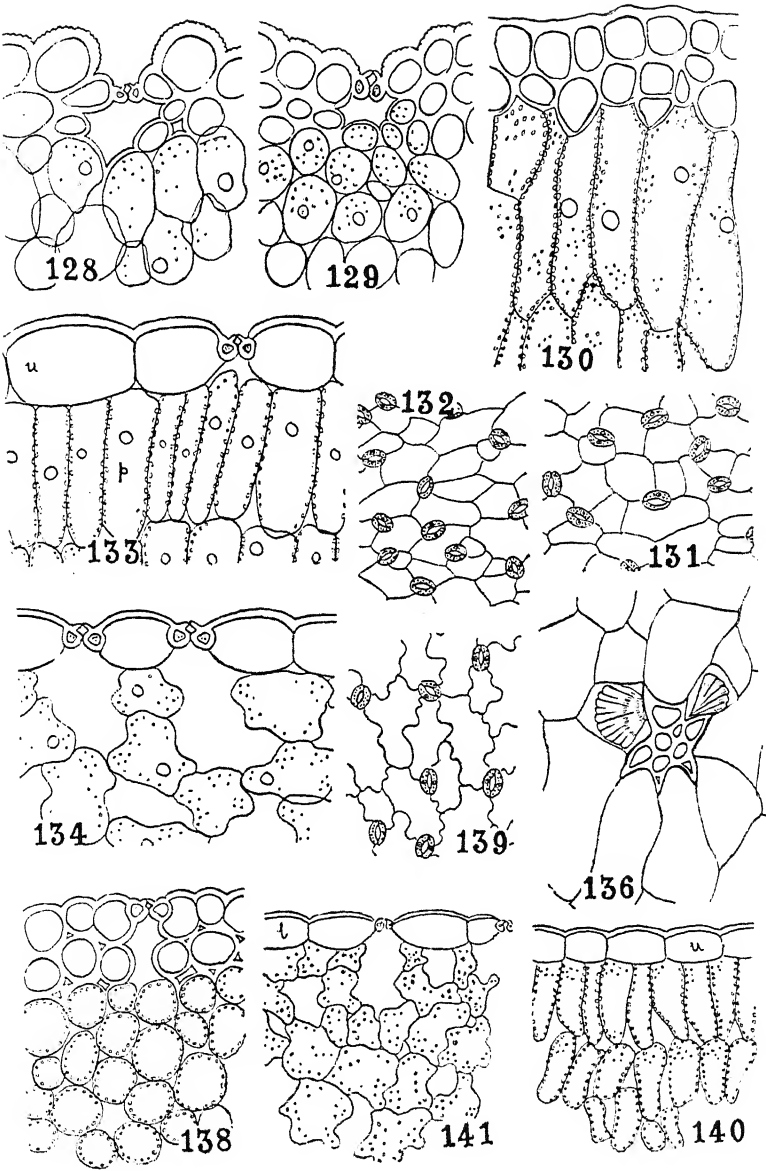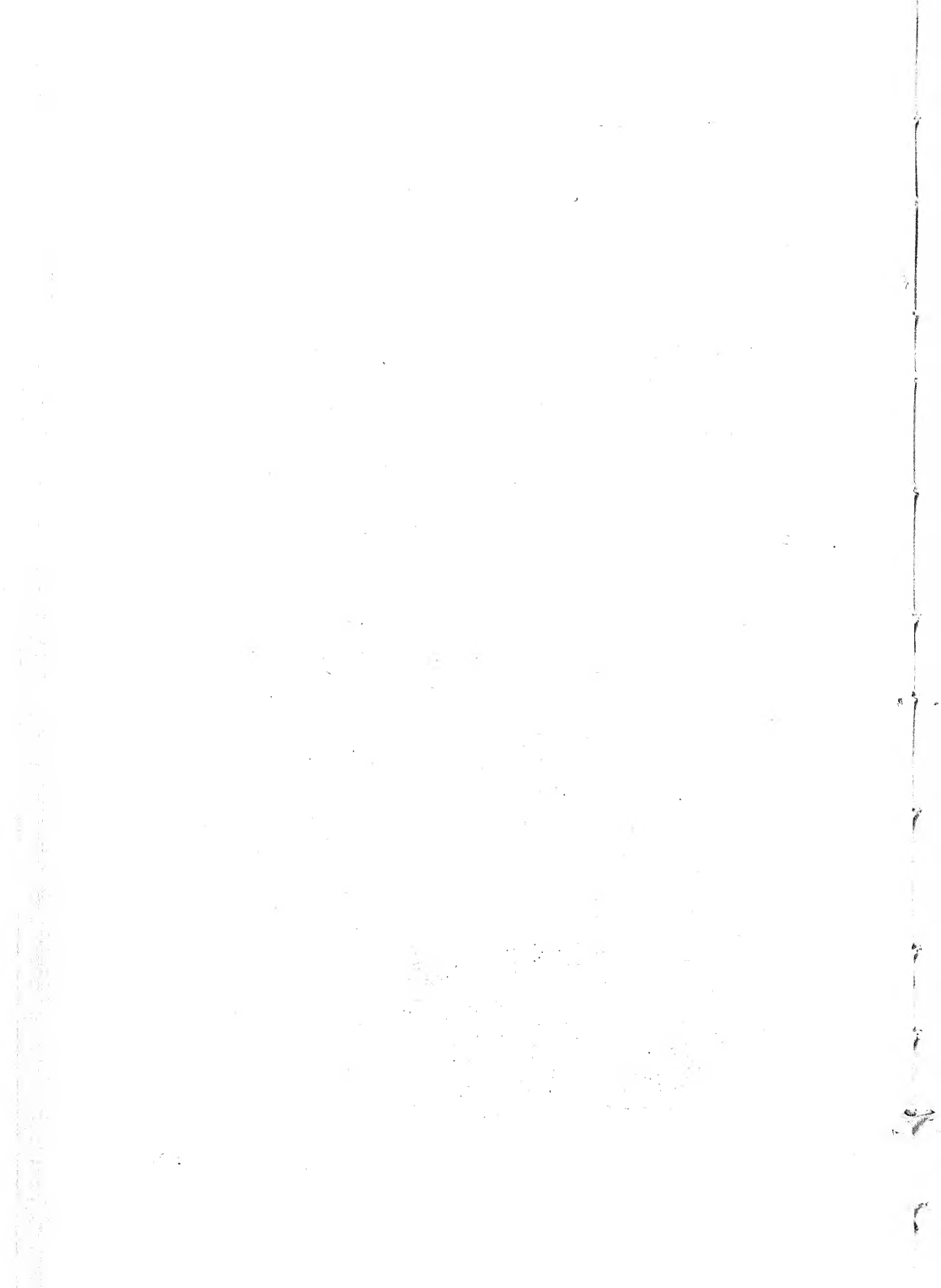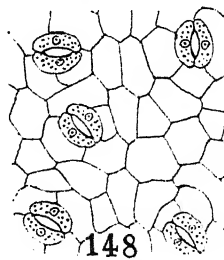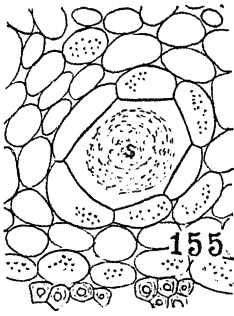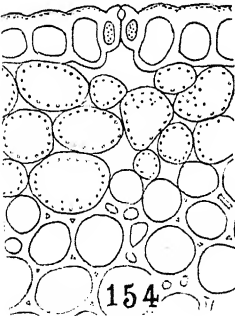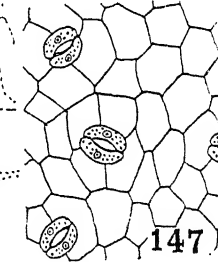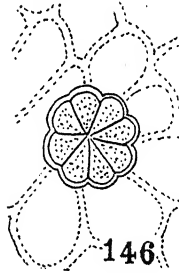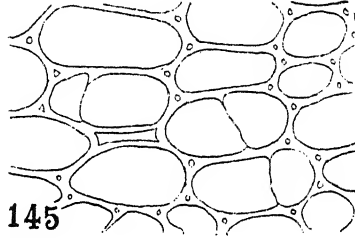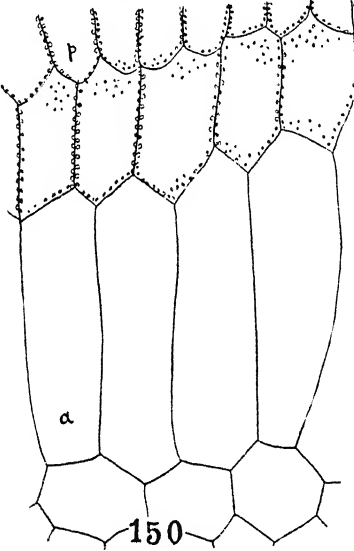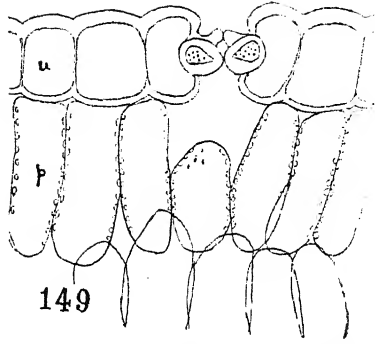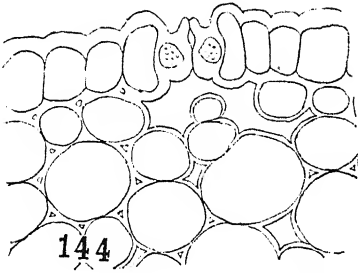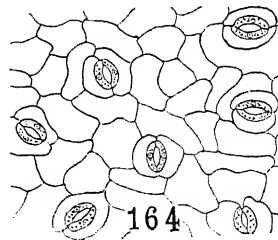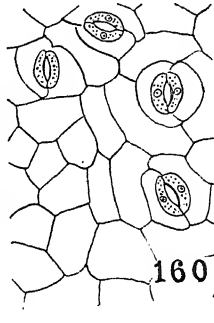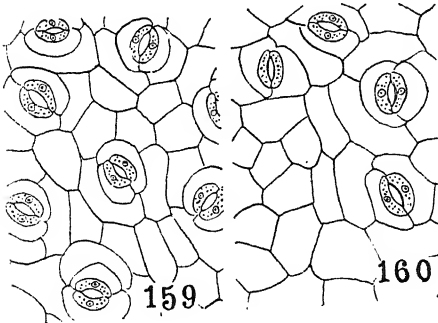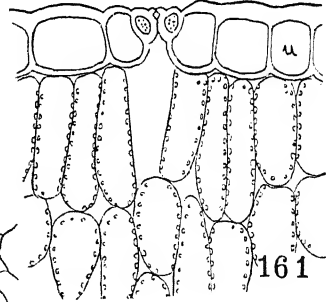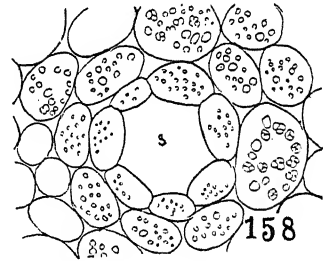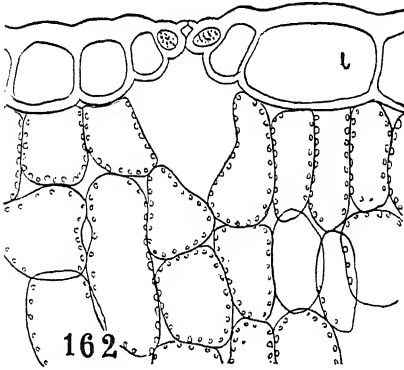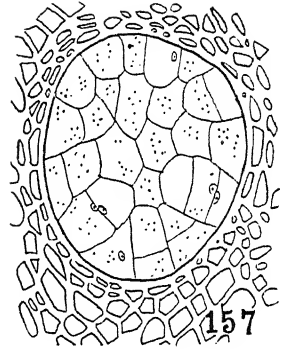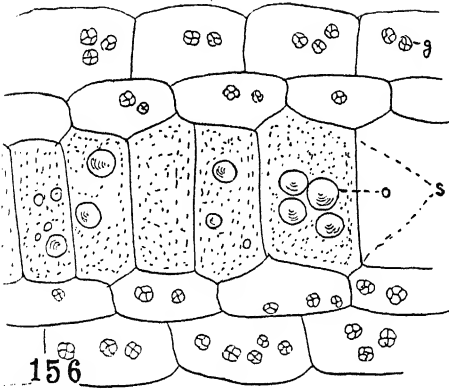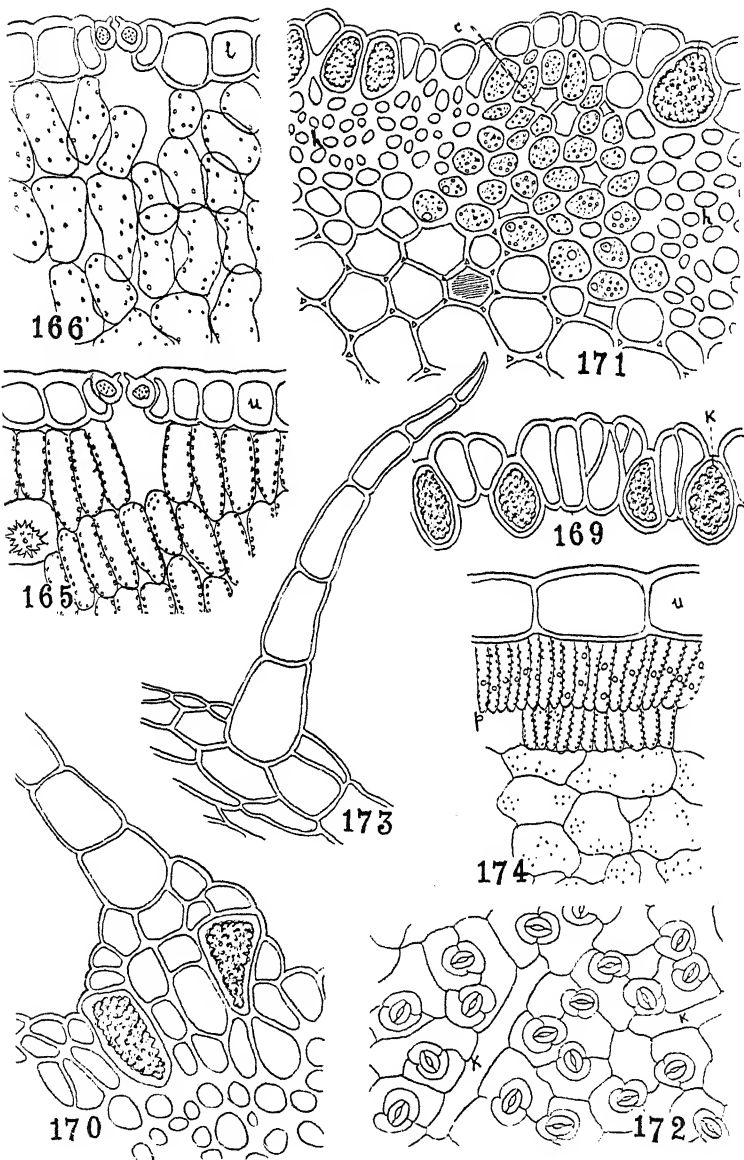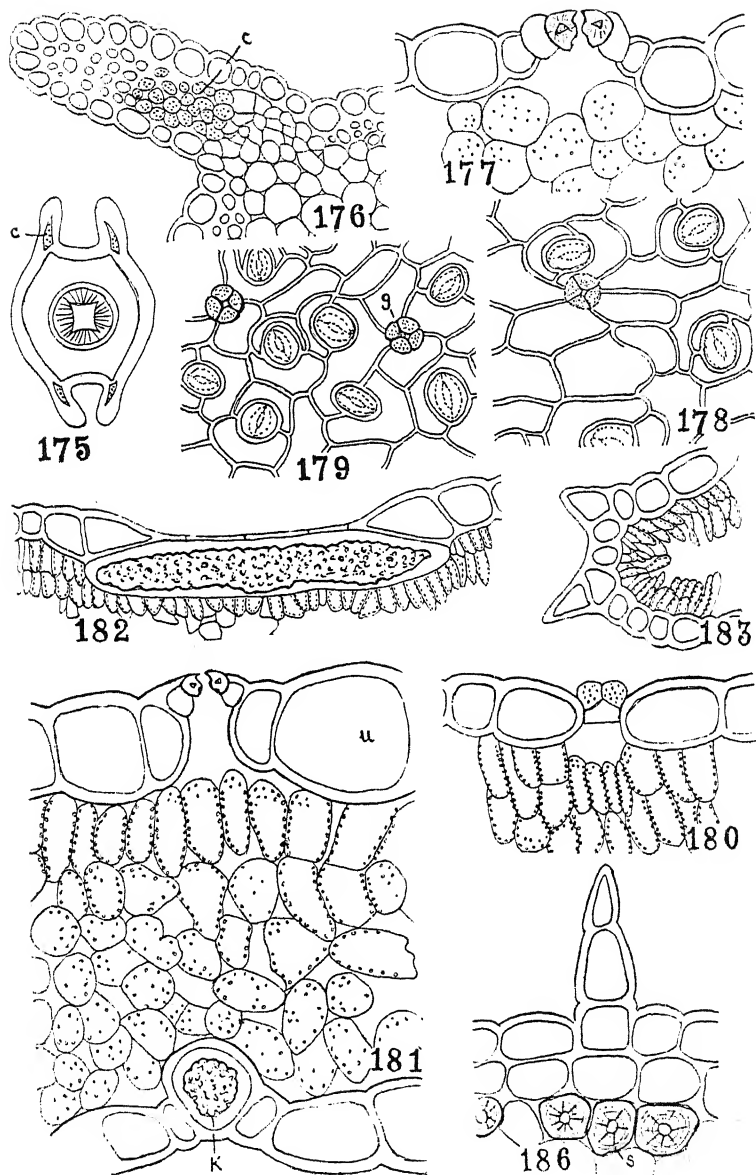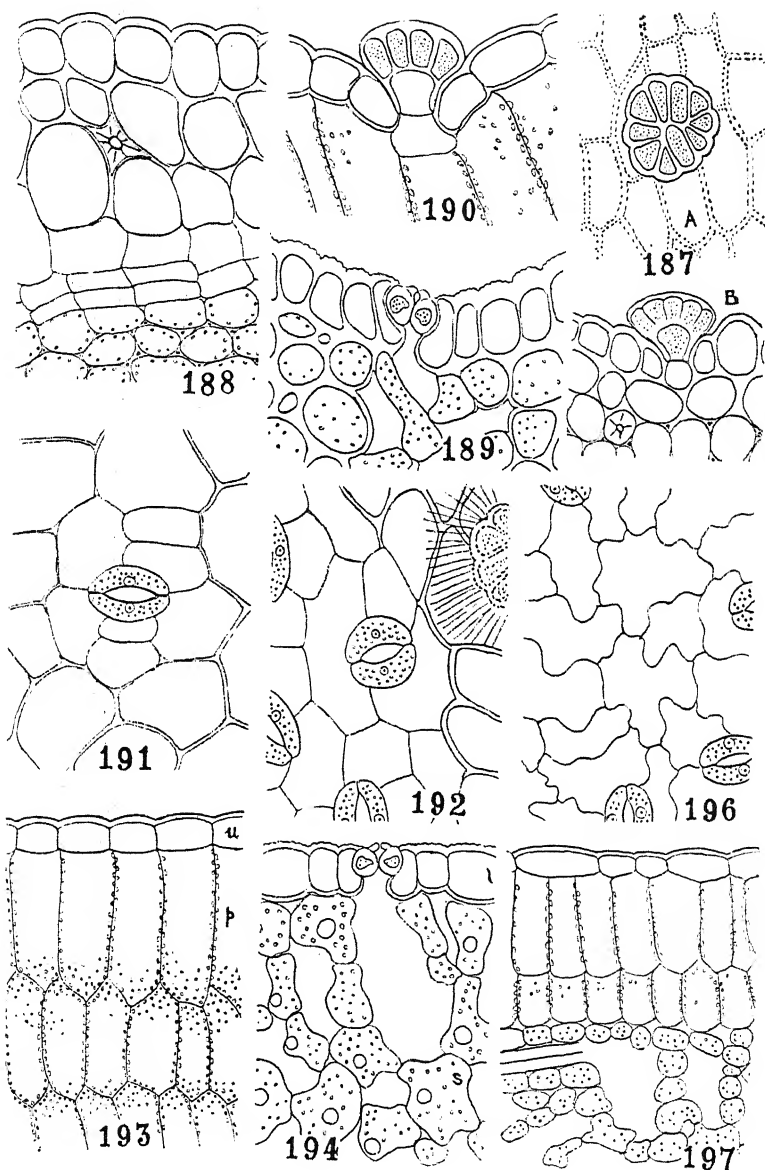
CHANGES IN PLANTS DURING LOW TEMPERATURES.

II. Is Hardiness [1] Inherited in Pyrus Malus?

BY

R. C. MALHOTRA,

Malhotra Street, Jammu, Kashmir.

Hardiness has attracted the attention of botanists and horticulturists for a long time, not only in apple but in other plants grown in the temperate climates. This is probably because freezing injury, at times, is so severe, at least in the temperate climates, that fruit growers suffer heavy loss both immediate and remote.

Plant physiologists and biochemists attribute hardiness to the relative amount of chemical substances present in a given plant. Rosa (25) and Hooker (14) are of the opinion that hardening is due to the increase of pentosans in a tissue or a plant. Murneek (21) suggests that hemicelluloses may be concerned in hardiness. On the other hand, De Long (4) and Doyle and Clinch (7) find no relationship between pentosans and hardiness in apple and conifers. Murneek also presents no direct experimental data in favor of hemicellulose.

Several physical or chemo-physical factors responsible for hardiness have been proposed. D'Arsonval (2) thinks that hardened plants have lower freezing point. Harvey's (11) experiments indicate a direct relationship between alkalinity and hardiness or acidity and freezing injury. Shutt (29) and others find that decrease in moisture content has a definite relationship with hardiness.

There is an indication that variation in the histological structures of the plant cells may be more important in hardiness than chemical or chemo-physical composition. Molisch (20) records that compact cells may withstand lower temperatures. Hursh (15) finds that in some cases morphological factors may be important. Woolsey (28) mentions that thickness of the bark may be an important factor.

Are all these factors interrelated? Can they be inherited? Conclusive evidence is not available in this respect. It seems that resistance and susceptibility are as stable as other genetic characters

---

[1] " Hardiness " as used in this paper refers to the ability of a given plant species or variety to successfully withstand the lowest range of temperature as well as freezing and thawing, snowfall and wind. Hardy plants have been designated for a geographical region based on the sum total of all the major environmental factors involved and not on the prevailing temperature only.

and may depend upon genetic factors. For instance, Hayes (12) found that inherited wheat varieties may be resistant to stem rust because of a large amount of sclerenchyma cells. Hayes and Aamodt (13) and Quisenberry (23) also found spring growth habit of wheat and less cold resistance as dominant over winter growth habit and cold resistance. In other words, cold resistance and winter habit of wheat are strongly correlated in inheritance. Waldron (26) concludes that hardiness is primarily inherent in the plant itself.

The problem of hardiness with reference to inheritance, be it due to the chemical, chemo-physical, anatomical, morphological or other factors not yet known, is not a simple one. The winter hardiness appears to be controlled by several genetical factors, the final expression being greatly influenced by the environment under which the material is grown, as for instance, growth habit. The winter-hardiness character, while heritable, is very complex and is greatly influenced by environment as shown by 10 years' experiments of Quisenberry and Clark (24).

Dorsey and Bushnell (5) pointed out that no cultural methods or protective treatments have been developed which are adequate to prevent serious injury when a variety is grown too far north of its normal range. Only origination of hardy varieties by breeding appears to be the effective procedure. Summarizing their (6) work on the inheritance of hardiness in plums, they state that from one standpoint the environment may be regarded as the primary consideration in hardiness and the genetic constitution secondary, or vice versa. Darwin (3) mentioned that Hooker found seedlings of the same species, grown from seeds at different heights on the Himalayas, to possess different constitutional powers of resisting cold.

The writer is interested in this problem both from a genetical and a biochemical standpoint. It seems that both morphological and chemical factors are hereditary to a greater extent, although environment can modify them. For instance Malhotra (19) found that tomato grown from the same parent of F_{10}, can be modified both as to the morphological structure and the chemical composition. The last fact has been illustrated by plate I.

Beach and Allan (1) conclude that apple varieties, varying in hardiness, are morphologically indistinguishable. The writer obtained Winter Banana (tender apple variety) and McIntosh (hard apple variety) from various localities of temperate regions (United States of America and Canada) and found no difference in the structure of wood of the same year except, that one had more tracheæ per unit surface than the other. Plate II shows the structure of the typical wood of these varieties of apple. It seems that these morphological similarities

or differences, are inherited.   On the other hand, hardy apple varieties, in general, possess thicker bark as can be seen from plate II.   This thickness is again influenced by environment, but at the same time, resistance against winter injury is also decreased.   Plate III shows the bark cells in both kinds of apples.   Thus it would seem that one part of the plant may be more influenced by inheritance while another may be more affected by environment.   Malhotra (17, 18) has already indicated that sex can be influenced by external factors.

It seems that apple varieties from the coldest regions are the hardiest when planted in the same locality.   The writer found this true for apple, while Emerson (9) and Hansen (10) found for black walnut seedlings, red cedar and boxelder respectively.   On the other hand, Linton (16), Ellis (8) and Neilson (22) state, that seedlings obtained from the seeds from warmer climates, may be hardy in cold regions. This may be due to inheritance or successful adaptation or both.   At any rate, it is quite possible that inheritance may be an important factor in securing hardy plants.   Several cultivated species have given rise to single individuals that are able to endure considerably lower temperatures than the individual types.   Many examples of this type have been noted by White (27).

It is very likely that wild, hardy varieties of apples exist. Investigation for discovering mutants or breeding hardy plants may be worth trying.   The solution of this problem does not lie so much in chemical and physiological attacks as in the hereditary basis of hardiness.   Unfortunately this phase has not attracted the attention it deserves.   The writer's researches will appear in Norway regarding the critical problems of this sort.

Literature Cited.

1. BEACH, S. A. AND ALLEN, J. W. JR.—Hardiness in the apple as correlated with structure and composition. Iowa Agr. Exp. Sta. Res. Bul. 21, 1915.

2. D'ARSONVAL M.—Le pression osmotique et son rôle de defense contrale froid cellule vivante. Comp. Rend. Acad. Sci. Paris. 133 : 84–86. 1901.

3. DARWIN, C.—The origin of species. pp. 133–34.   1859.

4. DE LONG, W. A.—Pentosan content in relation to winter hardiness in apple. Sci. Agr. 8 : 512–523.   1928.

5. DORSEY, M. J. AND BUSHNELL, J. W.—The hardiness problem. Proc. Amer. Soc. Hort. Sci. 17 : 210–224.   1920.

6. DORSEY, M. J. AND BUSHNELL, J. W.—Plum investigations II. The inheritance of hardiness. Minn. Agr. Exp. Sta. Tech Bul. 32. 1925.

7. DOYLE, J. AND CLINCH, P.—The pentosan theory of cold resistance applied to conifers. Sci. Proc. Roy. Dublin Soc. 18 : 219–235. 1926 ; also Proc. Roy. Irish Acad. Sci. Sect. B (37) 26 : 373–414. 1927.

8. ELLIS, Z. H.—The farthest north U. S. Nut culture. 16th Ann. Report Northern Nut Growers' Assoc. p. 29. 1925.

9. EMERSON, R. A.—The relation of early maturity to hardiness in trees. 19th Ann. Report. Nebr. Agr. Exp. Sta. pp. 101–100. 1906.

10. HANSEN, N. E.—The breeding of cold-resistant fruits. Report 3rd. Internat. Conf. Gen. pp. 401–404. 1906.

11. HARVEY, R. B.—Hardening process in plants and developments from frost injury, Jour. Ag. Res. 15 : 83–112. 1918.

12. HAYES, H. K.—Inheritance of disease resistance in plants. Amer. Natur. 64 : 15–36. 1930.

13. HAYES, H. K. AND AAMODT, O. S.—Inheritance of winter hardiness and growth habit in crosses of marquis with minhardi and minturki wheats. Jour. Agr. Res. 35 : 223–236. 1927.

14. HOOKER, H. D.—Pentosan content in relation to winter hardiness. Proc. Amer. Soc. Hort. Sci. 17 : 204–207. 1920.

15. HURSH, C. R.—Morphological and physiological studies on the resistance of wheat to *Puccinia graminis tritici* Erikss and Henn. Jour. Agr. Res. 27 : 381–411. 1924.

16. LINTON, W. A.—Roadside planting. 16th Ann. Report Northern Nut Growers' Assoc. p. 23. 1925.

17. MALHOTRA, R. C.—The sex ratio in Asparagus officinalis L. and its artificial modification. Jour. Genetics, 23 : 157–172. 1930.

18. MALHOTRA, R. C.—Can sex ratio be altered in diœcious plants ? Amer. Natur. 64 : 470–473. 1930.

19. MALHOTRA, R. C.—Hereditary mechanism of winter hardiness in some horticultural plants. Jour. Amer. Bot. Soc. (In press) 1932.

20. MOLISCH, H.—Untersuchungen ueber das erfrieren der pflanzen. Jena. 1897.
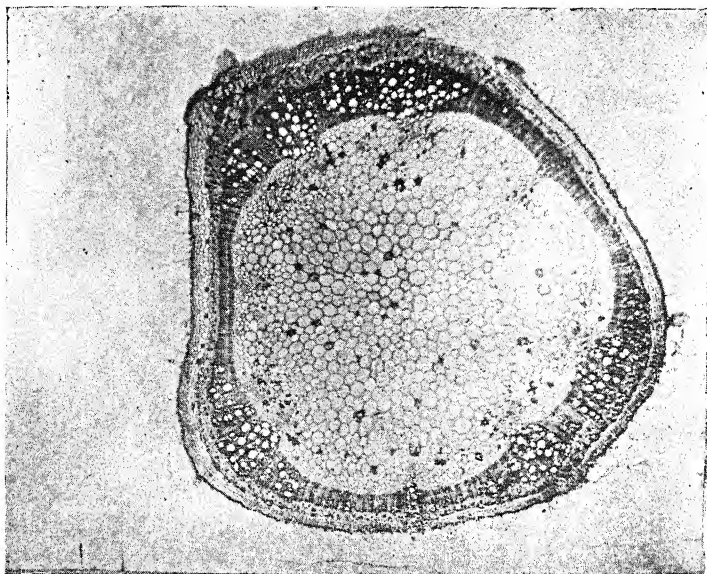
PLATE I.
Photomicrographs of the cross-sections of tomato plants obtained from homozygous seeds of F_{10}.
   A. Grown at 75^0 F. Note that the cells (particularly pith parenchyma), on the whole, are larger in size.
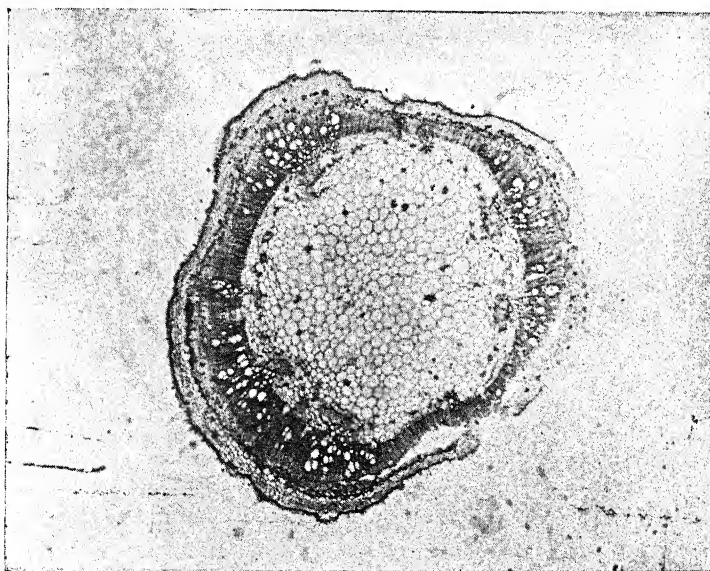
   B. Grown at 50^0 F. The cells, as well as the total cross-section area, are smaller than A.   This dimensional difference is probably due to environment.
   Dark areas in both A and B are due to iodine accumulation.
   The following tissues can be seen from outside to inside :—Epidermis with hair, collenchyma, pericycle, external phloem, xylem also medullary rays, internal phloem and pith. (Magnification X 500)
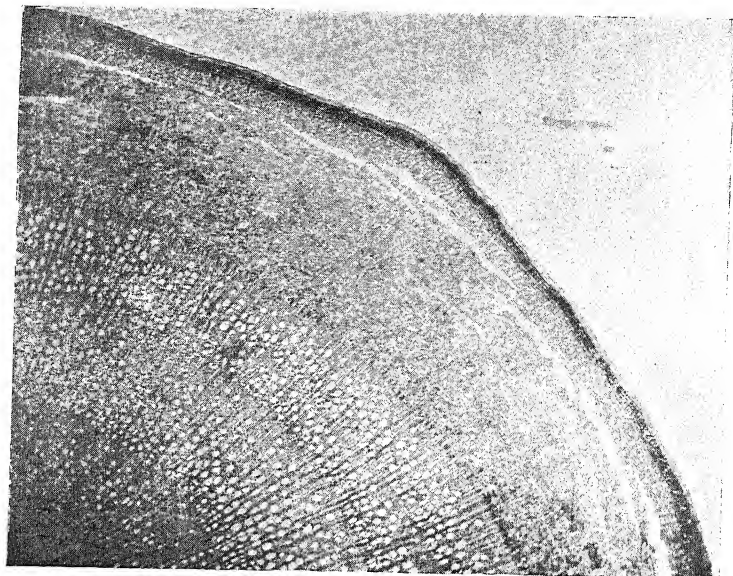
PLATE II.
Photomicrographs of the cross-sections of 2 years' Winter Banana (tender apple) and McIntosh (hardy apple) shoots.
A. McIntosh. Note similar cells in wood as that of B. except that they are fewer per unit area in B. On the other hand note the thickness of the bark in favour of A.
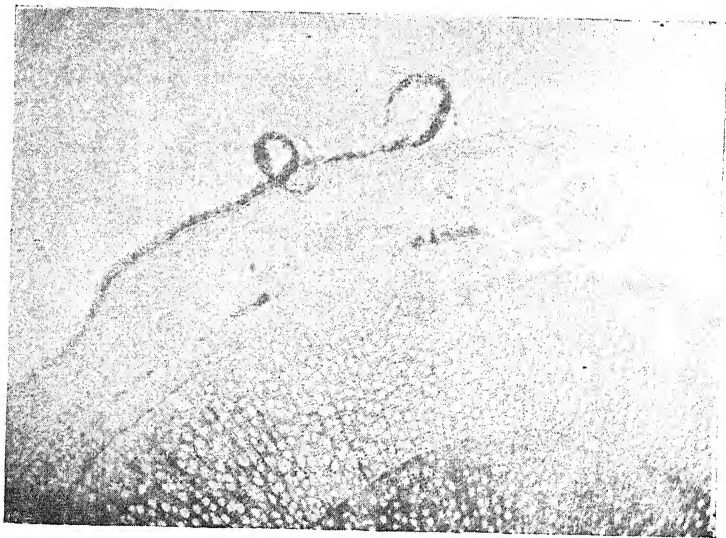
B. Winter Banana. Note the thinner bark than that of A. These two twigs were cut at the same distance from the tip and represent a typical structure of about 60 twigs collected from different trees grown in various localities. (Magnification X 500).

21. MURNEEK, A. E.—Hemicellulose as a storage carbohydrate in woody plants with special reference to the apple. Plant. Physiol. 4 : 193–212. 1929.

22. NEILSON, J. A.—The nut flora of Canada. 16th Ann. Rpt. Nut Growers' Assoc. p. 63. 1925.

23. QUISENBERRY, K. S.—Inheritance of winter hardiness, growth habit and stem-rust reaction in crosses between Minhardi winter and H-44 spring wheats, U. S. D. A. Tech. Bul. 218. 1931.

24. QUISENBERRY, K. S. AND CLARK, J. A.—Breeding hard red wheat, for winter hardiness and high yield. U. S. D. A. Tech. Bul. 136. 1929.

25. ROSA, J. T. JR.—Investigations on the hardening process in vegetable plants. Mo. Agr. Exp. Sta. Res. Bul. 48, 1921.

26. WALDRON, C. B.—Factors in hardiness. Trans. Iowa Hort. Soc. 53 : pp. 115–119. 1918.

27. WHITE, O. E.—Mutation, adaptation to temperature differences and geographical distribution in plants. Brooklyn Bot. Gard. Contr. 53. 1928.

28. WOOLSEY, T. S.—Studies in French Forestry. P. 282. 1920. John Wiley and Sons, N. Y.

29. SHUTT, F. F.—On the relation of moisture content to hardiness in apple twigs. Proc. and Trans. Roy. Soc. Canada, 9 : 149–153. 1903.

REVIEWS.

Heilborn, Otto, Studies on the Taxonomy, Geographical Distribution and Embryology of the genus *Siparuna* Aubl. Svensk. Bot Tidsk. 25: 202–228. 1931.

The author first gives a taxonomic account of the genus *Siparuna* (Monimiaceæ) with the distribution of the species. Two new species are also described. There is a tendency towards endemism in the genus.

The most peculiar thing is the development of the embryosac which the author has studied in *Siparuna Eggersii*. Each carpel contains a single anatropous ovule with a single massive integument. The nucellus is large and several (4–6) archesporial cells are differentiated. They divide periclinally to form the primary wall cells and megaspore mother cells. The latter, on reduction, produce tetrads of megaspores. During this time a hypostase of thick-walled cells differentiates in the chalazal region of the nucellus. The upper three megaspores of each tetrad now degenerate while the chalazal megaspores grow out into long and narrow tubes penetrating downward into the nucellus. Usually only the tube formed by the megaspore from the central tetrad continues to grow; the others are crowded out and absorbed. This tube soon encounters the hypostase. Here it swells into a small vesicle and as it is prevented from stretching further by the cells of the hypostase, it becomes coiled round itself. The megaspore nucleus and its protoplasm gradually wander into this part. Meanwhile, some of the nucellar tissue above the hypostase disintegrates and a cavity is formed into which the embryosac-tube protrudes and discharges its nucleus and protoplasm. The cavity increases in size and the megaspore nucleus either does not divide at all or at the most 5 nuclei are formed without any organisation of the egg apparatus. Later, all these degenerate. The nucellar cavity enlarges still further and in its micropylar part a small sporophytic bud arises, but never develops beyond the three-celled stage and finally becomes destroyed and absorbed. The fruits are always well developed, but the seeds are shrivelled, empty and sterile.

The peculiar mode of development of the embryosac described in this paper by HEILBORN has not been reported before in any other plant. It will be interesting to know the condition in other plants of the same family.

P. MAHESHWARI.

Chamberlain, C. H.—Methods in Plant Histology. Fifth revised edition. 1932. 416 pp. University of Chicago Press. $3·25.

The fourth edition (1924) of this very useful book has again been revised and brought up to date. It has been the author's constant endeavour to present his "Methods" in such a way that not only experienced technicians but even students can profit by reading the book.

There are additions, alterations and improvements in almost every chapter, so that about 70 new pages have been added.

The chapter on "Introduction" has been almost entirely re-written. The author advises everyone who works with a microscope to try to be a good technician and emphasizes that the making of slides should not be entrusted to an assistant. "Those who think that such work is mere mechanical drudgery, which can be done by an assistant, are likely to become armchair investigators, drawing false conclusions or becoming scholastic grafters, according as the assistant is mediocre or talented."

Some new suggestions and combinations are given in the chapter on "Stains and Staining". The "Paraffin method" has been further improved and suggestions are given for cutting sections as thin as 1 micron and even less. Instead of going to the drudgery of constantly sharpening microtome knives, the author favours the use of suitable safety razor blades clamped in blade holders. In chapter XII, there are two important additions in the methods for cutting sections of hard wood—KISSER's Steam method, and JEFFREY's Vulcanising method. There are two new chapters on "Palæobotanical Microtechnique" and "Illustrations for Publications". The chapter on "Botanical Photography" has had many additions and improvements and parts are written by Dr. Paul J. Sedgwick, an old student of Prof. Chamberlain.

The II Part of the book is devoted to specific directions on the collection, preservation and study of important representatives from the whole range of the plant kingdom. A list of references is also given and the author expresses the belief that "an ambitious student by making preparations, studying them, and reading about them, can become well grounded in comparative morphology without attending any classes". The Bibliography is fuller, though some important papers on microtechnique are still left out. Among others, there is no mention of TAYLOR's Smear Method and TUAN's new method of differentiating Haematoxylin.

The get-up of the book is excellent and like the previous editions it will be indispensable to workers in Plant Morphology and Cytology.

P. MAHESHWARI.

A HANDBOOK OF SOME SOUTH INDIAN WEEDS

BY

C. TADULINGAM, F.L.S., AND G. VENKATANARAYAN, B.Sc. (AG.).,

Superintendent, Government Press, Madras, 1932.

The book consists of Introduction and six chapters dealing with

(1) The parts of a plant

(2) Nomenclature of plants.

(3) The weeds and their classification

(4) Special weeds

(5) Loss caused by weeds and their uses

(6) How weeds spread and the methods of controlling them, and description and notes on weeds. At the end a short Bibliography and a Glossary of technical terms used in the book are given. There are indices of Botanical Names, Common English Names, Tamil Names, Malayalam Names and Telugu Names. 112 species are described in the book, and the descriptions are supplemented by many interesting and useful observations. The book is fully illustrated and ought to prove very useful to all concerned. The printing, paper and binding are all very satisfactory.

LAHORE,

6th February, 1933.

STUDIES ON *ISOETES COROMANDELINA* L.
I. Sporogenesis.

BY

T. EKAMBARAM

AND

T. N. VENKATANATHAN.

Introduction.

The genus *Isoetes* has been the subject of repeated investigations in the past.[*] Ever since the time of Hofmeister the structure of the adult sporophyte attracted the attention of several workers, partly because of the peculiarities of structure and partly owing to the comparative ease with which it could be handled. Still the morphology of the tuberous axis is a difficult and fascinating problem. Moreover, the developmental stages of the sporangium and the embryo are less known on account of the difficulties in the methods of study and perhaps also due to the scarcity of material. Hence our knowledge of the life-history of the genus is by no means complete.

While the object of the present work was to follow the complete life-history of the local species, the sporangial structures were first taken up for investigation as the information available on these parts of the plant was most meagre. It is the purpose of this paper to describe in detail the development of the sporangia, which, while adding to the present knowledge of the life-history of the genus, may also be useful in elucidating the phylogenetic relationships of the genus.

---

[*]For a complete list of papers on *Isoetes* to date refer to H. Liebig's paper "Erganzugen zur Entwicklungsgeschichte von Isoetes lacustre L. Flora. Bd. 125. 1931.

Review of Previous Work.

The first clear account of the origin and early development of the sporangium was given by Hofmeister (15). According to him the sporangium and the velum have a common origin from a single superficial cell situated underneath the place of insertion of the scale (the ligule). This divides by a transverse septum into two cells, the upper one becoming the primary cell of the velum while the lower becomes the primary mother-cell of the sporangium. The latter, by repeated division in all directions, forms the sporangium which at first consists throughout of homogeneous thin-walled cells. The development proceeds alike in both kinds of sporangia up to certain stage, when the differentiation into three different kinds of tissue takes place. He has also given a few observations on the division stages in the megaspore mother-cells wherein he sees "two flatly spherical accumulations of granular matter" just before the nucleus vanishes, the presence of an evanescent interkinetic cell-plate, and the division of the mother-cells into bilateral quadrants.

Hegelmaier (16) and Tschistiakoff (32) traced the sporogenous tissue to a mass of deep lying meristem between the epidermis and the vascular bundle. The wall of the sporangium is described as being clearly delimited in the early stages from the inner complex which gives rise to the sporogenous tissue. After a careful study of *Isoetes lacustris,* this view was elaborated in detail by Goebel (12) whose account forms the basis of all text-book descriptions. The important facts of this description are: (1) the sporangium arises mainly from three outer layers of cells. (2) there is an hypodermal archesporium which gives rise to all spore mother-cells, trabeculae and tapetum. (3) each of the archesporial cells exhibits an independent growth and up to this stage the development of the microsporangium and the megasporangium is the same. (4) the further development of the two kinds of sporangia differs in their manner of growth, the megasporangium being marked by the absence of anticlinal divisions.

These observations have been generally confirmed by subsequent authors as Sadeback, Farmer, Schenck and others. The next important account on the development of the sporangium was given by Bower (4) who adds certain details to Goebel's description and generally confirms his account. But he differs from Goebel in one important point in that he traces the sporangium

to a transverse group of superficial cells. Thus he brings *Isoetes* in line with the other Pteridophytes with respect to the superficial origin of the archesporium. His primary object in working out the development of the sporangium was to trace a nearer affinity of the genus to the Lycopodiales.

Wilson Smith (36) working on *Isoetes engelmanni* and *I. echinospora,* confirms Bower's results as to the origin of the sporangium with variations in minor details and attempts to explain Hofmeister's statement as due to his exclusive dependence upon longitudinal sections. But as to the later stages of development, he entirely disagrees with the earlier authors. The sporangium, as described by him, consists throughout of a uniform mass of cells and he does not find any difference in the archesporial cells, either in their manner of development or their mode of growth. There is no single definite hypodermal archesporium, certain of whose cells have an independent growth. The spore-mother-cells are not morphologically predetermined but physiologically selected from among the large number of potentially sporogenous cells. The sporangium is single and not multiple and grows as one unit. He has besides studied in detail the further development of the microsporangium. He describes clearly the details of development of the trabeculae and the tapetum bud did not attempt to follow closely the details of the reduction divisions. His limited observations on this subject refer to (*a*) the chromatin passing through a synapsis stage, (*b*) the polycentric origin of the achromatic figure and (*c*) the divisions being of the simultaneous or successive types resulting in bilateral or tetrahedral spores respectively.

Fitting (11) followed the development of the megaspore mother-cells in the living material of *Isoetes lacustris* and *I. Purieui.* He gives a clear account of the formation of a body in the cytoplasm which divides twice prior to nuclear division. Of the latter, he describes the anaphase of the first division and the second division giving figures for the same.

The latest work is that of Ekstrand (10) who mainly supplements Wilson Smith's work on the microsporangium. He is of opinion that the sporangium grows as a number of "blocks" and not as one unit as maintained by Wilson Smith. He notes a few stages in the reduction division of the nucleus and observes that the division is marked by the absence of centrosomes or centrosome-like bodies. The chromosome number recorded by him for the species was 11.

It will be seen from the above summary that while the earlier history of the development of the sporangia received greater attention in the hands of the several authors, the later stages have not been studied in as much detail and the cytology of the reduction divisions received almost no attention.

Material.

Isoetes coromandelina, the only species so far recorded in India, grows very commonly on the Coromandel Coast. It has also been reported by Griffith (3) from Serampore in Bengal. It is perennial like all other species, with a distinct growing season which starts about mid-October after the beginning of the monsoon and continues till March. It is amphibious in habit, fringing the water edges of lakes, ponds or pools. It is never found in deep water, a portion of the leaves being above the water level.

After a preliminary study of the material collected during January 1930, it was followed throughout the next season, material being collected at regular intervals up to the end of the season. One locality was chosen for close study and the observations made here were generally confirmed by those in other localities in and around Madras.

The collection of microsporangia was a matter of great difficulty. There is no published record of the presence of microsporangia and microsporophylls in this species. Pfeiffer (22, p. 110) who obtained the material from India observed a few microspores 'distinctly recognisable as such scattered among the bases of sporophylls, sometimes in small groups.' But she found all the mature sporangia bearing only megaspores. Our preliminary observations generally confirmed these statements, and following the version usually given in text-books attempts were made in vain to collect microsporangiate material at the end of the season. But very careful search made again throughout next year resulted in our finding a few specimens with microsporangia. Most of the plants produce only megasporangia while plants forming microsporangia were less common. In such of the latter plants the microsporangia appear very early in the rainy season. The proportion of bisporangiate plants to megasporangiate can be roughly estimated to be 2 or 3 to 50. There was no external difference between the megasporangiate and the bisporangiate plants.

As to the succession of sporophylls in the species, the first two or three leaves that appear as soon as the resting corm starts

growth are either sterile or possess only abortive sporangia and the later ones are all fertile. In the megasporangiate plants all the sporophylls bear megasporangia only. In the bisporangiate plants, usually one or very rarely two microsporophylls have been observed to develop. They usually come up after two or three megasporophylls are formed. In those cases where two microsporophylls are formed in a single plant, the second one does not follow immediately the first one but comes up later on after a few megasporophylls have been formed. In the megasporophylls that are formed towards the end of the growth period the spores do not reach maturity, their full development being probably arrested by the onset of the dry season.

It may be interesting in this connection to note the arrangement of the sporophylls in some of the other species of the genus. In *Isoetes lacustris*, the megasporangia are situated on the outer, and the microsporangia on the inner leaves of the rosette, while the innermost zone is made up of sterile leaves. Campbell (8, p. 438) and Milde (21, p. 245) found that the outer leaves of each cycle produced microspores. Glück, (13) who made a special study of the biology of *Isoetes* observed numerous variations in the arrangement of the mega- and the micro-sporophylls in the water and land forms of several species investigated by him. His tables are very interesting and some species show no regular sequence in the formation of the sporophylls. Wilson Smith (36, p. 323) has also noted an occasional irregularity in the order of succession of sporophylls in the plants grown in his laboratory. "In *I. engelmanni* it was not at all uncommon to find several megasporophylls among the microsporophylls. Some plants which have been growing rapidly for seven or eight months in the laboratory formed only megaspores; some others, though producing a few microsporophylls failed to bring any microspores to perfection." West and Takeda (33, p. 334) observe in *Isoetes japonica* that sterile leaves form the transition from one year's increment to the next, and to occur on the periphery of the rosette. The mega- and the microsporophylls are not arranged in any definite order but irregularly distributed throughout the wide zone of fertile leaves. While stating that the general text-book version, (viz., that the megasporangia are situated on the outer leaves of the rosette, and the microsporangia on the inner, while the innermost zone is made up of sterile leaves) is incorrect, they seek to explain the error as follows: "when a plant is dug up late in the year it often happens that the sterile leaves of the outermost whorl

of the rosette have by that time completely decayed away; under these circumstances sporophylls are found on the margin. But if such a leaf-rosette be carefully dissected, young sterile leaves will be found in the centre; these are generally regarded as belonging to the present season's growth but actually represent the outermost circle of next year's growth''.

Methods.

As to the technique, the usual methods were employed no special methods having been found necessary for the study of the sporangiate material. While several fixatives were tried, stock chromo-acetic acid and Flemming's weaker solution generally gave the best results. A saturated solution of picric acid in water and an alcoholic solution of mercuric chloride with a few drops of acetic acid were also used with some success, though they were not found to be useful for the later division stages of the megaspore mother-cells. The fixation was partly hastened by an air-pump. The material was washed, dehydrated, and cleared in xylol in the ordinary way and was embedded in paraffin of 54° C. Sections were cut varying in thickness from 6–10 U. Haidenhain's iron-alum hæmatoxylin, saffranine and gentian violet, and picrocarmine in the case of material fixed in picric-acid and mercuric chloride were the stains used. While haematoxylin gave the best results others were also useful for some of the cytoplasmic structures.

In order to ensure the proper fixation of the megaspore mother-cells in the division stages, it was found necessary to cut open the sporangia and expose the sporangial tissue to the free action of the fixing fluids. Failing this precaution sections show the mother-cells plasmolised and contracted to a mere fraction of the original size which take a very intense stain so as to obscure totally the internal structure—a condition observed by Kienitz-Gerloff (16), Wilson Smith (36), Fitting (11) and other investigators.

The material which showed the division stages in the mother-cells was fixed both in the field and in the laboratory between 11 a.m. and 3 p.m. on warm sunny days.

Observations.

All observations made here are from permanent preparations of microtome sections. Some of them have also been confirmed by examination of fresh material. It will be convenient to start

with the description of the velum as it has been a structure closely associated with the sporangium. This will be followed by the general developmental history of the sporangium and, finally, the details of the reduction divisions in the mother-cells.

The Velum.

The velum is a scale-like structure, situated immediately below the ligule at its base and appears very early in the history of the development of the leaf. The degree of development of the velum varies for the several species, and this character, in conjunction with other prominent features, proves to be a fairly dependable diagnostic feature in the primary divisions of the genus. It takes its origin from the tissue between the ligule and the sporangium and possesses an upper and a lower lobe. The upper lobe—the 'labium' of A. Braun, grows in an upward oblique direction and has a very short growth. The lower lobe grows downward to a greater or lesser degree and is continuous with the tissues of the leaf at the sides. In a few cases it covers the whole sporangium like a pocket leaving only a slit opening below. This complete velum is characteristic of the terrestrial species, of *Isoetes*, e.g. *I. hystrix* (27, p. 426). In many other species, the development is not so complete, the lower lobe covering the sporangium only partially. In some others the lower lobe is totally absent. In *I. coromandelina*, only the upper lobe is developed (Fig. 6) and the lower lobe is not formed at all.

There has been much disagreement among the several investigators as regards the development of the velum. Most of them are of opinion that it takes its origin from the upper tiers of cells resulting from the divisions of the sporangium initials. While Hofmeister (15, p. 365) describes it as being separated from the sporangium by the very early divisions of the sporangium initials, others like Wilson Smith (36, 242) contend that it makes some addition to the sporogenous tissue. Anyway, it is taken as 'the sterilised portion of the sporogenous tissue' or 'a part of the sporangium fundament'. Braun (7) and Scott and Hill (27), however, could not find any relation between the development of the velum and the sporangium and they described it as having its origin from a distinctly separate tissue between the ligule and the sporangium—the 'sella' of Braun. Our observations are in agreement with those of the latter authors. The velum is far advanced in development before any trace of the sporogenous initials could be found below it.

The details of development of the velum in *I. coromandelina* briefly described are as follows: when the ligule has grown to a length of 6–8 cells the velum has already made its appearance as a three-celled structure just below the ligule which projects out
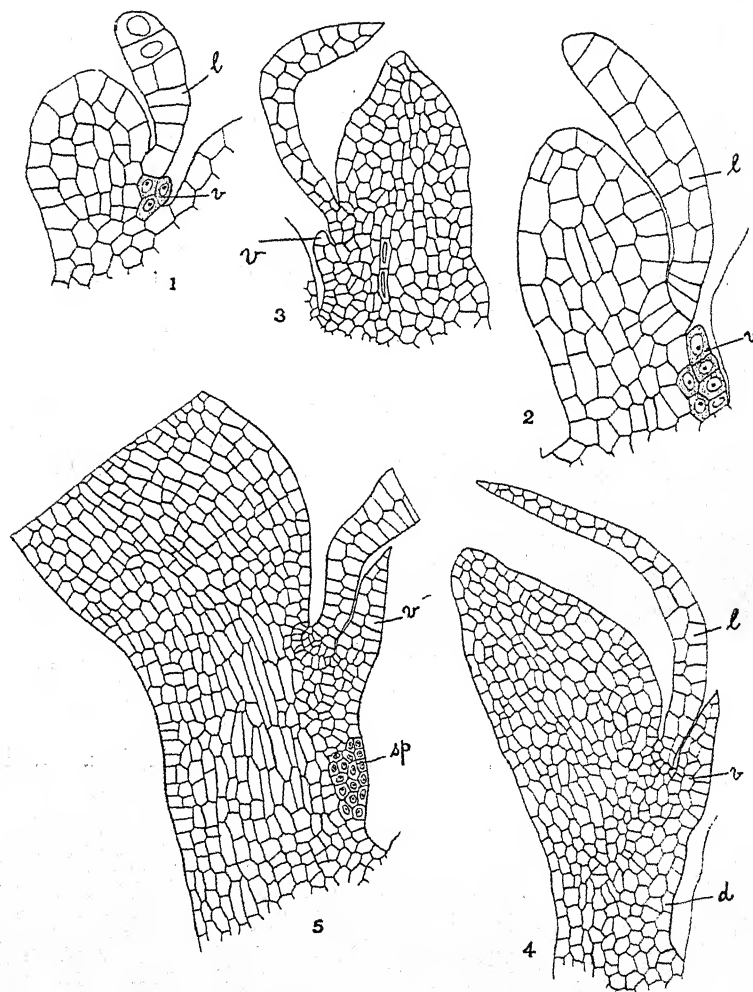
Figs. 1 and 2. Longitudinal sections of the leaf with the rudiments of the velum and the ligule (*l*). $\times$ 630. Fig. 3. Same; a slightly later stage. The velum (*v*) projects out in an oblique upwards direction. $\times$ 300. Fig. 4. Same; more advanced; (*d*) group of cells between the velum and the leaf-base from which arises later the sporangium. $\times$ 300. Fig. 5. Long. section of the leaf with the rudiments of the sporangium (sp.). $\times$ 300.

into the funnel-shaped cavity formed by the overlapping leaves in the young "bud" (fig. 1, v). This soon becomes a 5–6-celled structure and stands at an oblique angle with its free end upwards (fig. 2, v). As the ligule grows in size, the velum also keeps pace with it by a vigorous multiplication of its cells in all directions and continues to grow upwards in an oblique direction. At the same time, the tissue forming the base of the leaf below the velum produces a group of cells by active division (fig. 4, d). The sporangium takes its origin in the fertile leaves from this group of cells. The cells of the sporangium rudiment are distinctly recognisable by their rich contents sharply contrasting with the cells of the velum, which are relatively poor in their protoplasmic content, and appear only now for the first time in the history of the development of the leaf (fig. 5. sp.). They cannot be observed at an earlier stage than this. Even in the sterile leaves which are formed very early in the course of the vegetative season the velum is formed, only the sporangium is not developed from the group of cells below it and the base of the leaf below the velum remains very short. The subsequent growth by division of cells takes place in the region of the ligular base and the velum, by which the tissue of the ligule is differentiated into the sheath, glossopodium, etc., and the cells of the velum are enlarged. The growth of the leaf gradually shifts from its base to the portion above the ligule where it continues for a long time as an intercalary meristem. This gradual shifting of meristematic activity from the cells of the leaf-base to those above the ligule occurs in the fertile leaves also, after the sporangium has been well developed.

In this species the upper lobe of the velum is fully developed at a very early stage in the growth of the leaf. In its fully developed condition, it is a tongue-shaped structure distinctly delimited from the sporangium. (fig. 6). It has a clear epidermis at the free portion. The cells in the interior have grown very large and practically lost their contents. The most important characteristic of the velum is that many of the large cells of the interior possess a tracheidal structure with spiral or annular thickenings on their walls. The tracheidal structure is at first developed in the cells at the upper free portion of the velum and later on in the cells lower below.

The tracheids from an irregular network and are found in abundance near the ligular base and in the free portion of the

velum. They are comparatively few near the vascular bundle of
the leaf. Wilson Smith (36, p. 242) did not find any connection
between these tracheidal cells and the leaf trace in *I. echinospora*
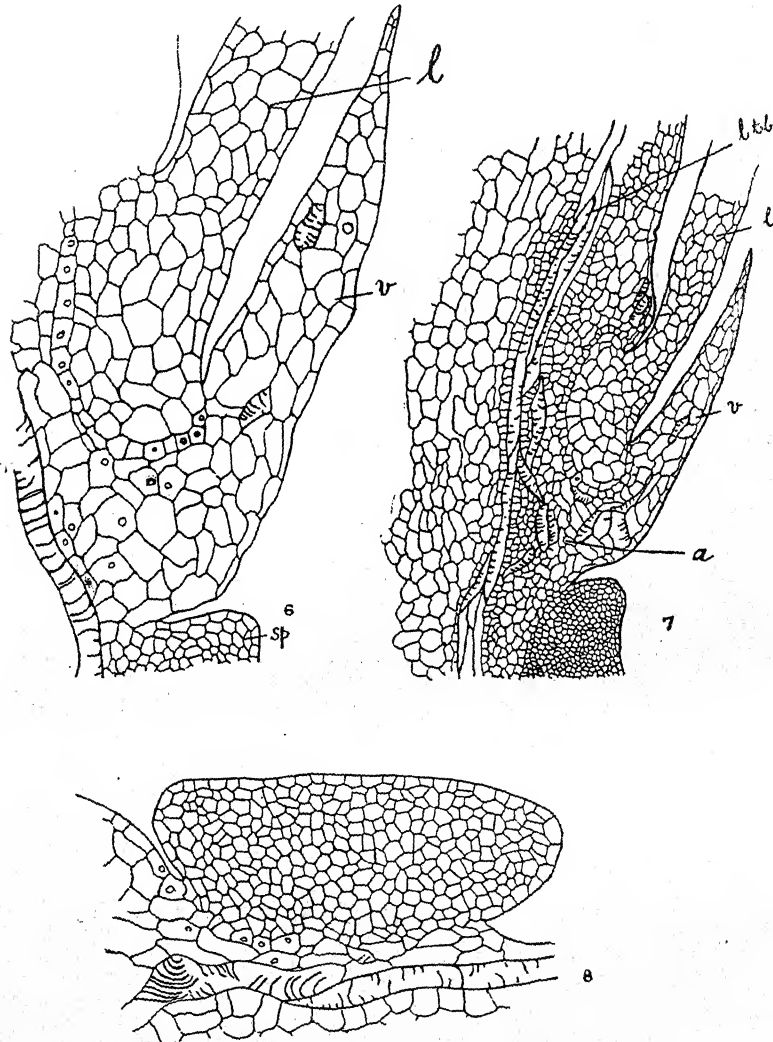
Fig. 6. The fully developed upper lobe of the velum. Fig. 7. A long.
section of the leaf in the velum portion showing the connection between the
leaf trace bundle and the tracheids of the velum. (ltb) leaf trace bundle,
× 150. In the next section of the series a tracheid lying in transverse
direction fills the gap (marked by cells at a) in the tracheidal connection.
Fif. 8. A young sporangium showing a uniform mass of cells. × 300.

and *I. engelmanni.** In the present species, however, we find a definite connection between the two by means of a narrow strip of tracheidal cells. (fig. 7). This connection is rather hard to detect at first but can be definitely made out by a careful study of the serial sections of the "bud".

The following are the chief points resulting from the above observations:—

1. The origin of the velum independent of and preceding the development of the sporangium.

2. The development and differentiation of the velum even in the sterile leaves where no sporangia develop.

3. The connection between the vascular tissue of the leaf and the tracheidal elements of the velum.

All these characters go to prove that the velum is a purely vegetative structure. The exact homologies and functions of this structure remain unknown for want of satisfactory evidence either from the fossil or living allies of the genus. The presence of the tracheidal network as a branch of the leaf trace bundle indicates that the velum, unlike the ligule which presumably is a later interpolated structure, is a more fundamental part of the leaf, which in the course of evolution has degenerated. The velum according to this view will represent a vestigeal structure.

The Sporangium.

(*a*) *Origin and early development:*—The sporangium arises from the upper surface of the sporophyll between the velum and the base of the leaf (fig. 5 sp.). As our observations on the developmental history of the sporangium are essentially similar to those of Wilson Smith (36), only the important points will be repeated here without any elaboration of detail.

The sporangium is superficial in origin, arising from the uppermost layer of cells of the leaf between its base and the velum. This layer of cells by successive divisions in all directions gives rise to a mass of sporangial tissue. The divisions do not follow any regular order of sequence and an epidermal layer as distinct from the inner archesporial mass could not be made out in the earlier stages. The outer layer of cells continues to add for some time to the inner mass of tissue by periclinal divisions. The

---

*Similarly, Scott and Hill (27, p. 435) state that the sub-ligular xylem may nearly reach the foliar bundle, but they have never observed actual continuity between the two tissues.

young sporangium is a mass of uniform cells and it grows as one unit. Usually the middle group of cells in the sporangium are more actively dividing in the earlier stages, but division figures of the nucleus may often be noted in the other regions also. Our preparations do not offer any kind of evidence in support of the archesporium theory of Goebel; the delimitation of sterile tissue during the very early stages into the several parts such as the epidermis, archesporium, etc., and the independent growth of each of the archesporial cells as described and figured by Goebel could not be observed by us.

When the sporangium has attained a fairly big size as that shown in fig. 8, changes take place in the uniform mass of sporogenous tissue which determine the nature of the sporangium. These changes are not similar in the mega- and the microsporangia and will now be dealt with under two separate heads.

The Megasporangium:—In the megasporangium, a few individual cells, or sometimes groups of two or three cells enlarge in size and are marked out from the neighbouring cells by their denser protoplasmic content and larger nuclei. Such cells or groups of cells form a discontinuous plate following the contour of the outer surface of the sporangium and lying below a mass of uniform tissue four or five cells deep. In section, they form a broken row at the same depth from the surface. But this arrangement is by no means the rule; these cells have been observed sometimes nearer the surface and at other times much deeper towards the inner wall of the sporangium. These cells are the potential megaspore mother-cells.

All the potential megaspore-mother-cells do not, however, reach maturity and give rise to the megaspores. Just after the differentiation of the megaspore mother-cells the sporangium passes through a period of enlargement. At this stage all the mother-cells show an increase in size becoming several times larger than the surrounding cells. But very soon growth in some of the mother-cells is arrested and their contents become less dense and somewhat vacuolated, with the result that the mother-cells in various stages of growth are met with. Besides this arrest in growth, there is also a widespread degeneration and abortion of many of the mother-cells which have reached their full size, marked by their divisions into smaller cells. The divisions in these may occur once, twice, or even three times, a group of eight cells also having been observed. Groups of 3, 5, and 7 cells are

not uncommon, suggesting that the division is not uniform in all of them. The resulting cells of each of these mother-cells do not fall away from one another, but are in groups retaining the contour of the mother-cell. Ultimately they attain the size of the cells of the sterile tissue. This widespread degeneration of the potential mother-cells has been dealt with in detail by Bower (5) to illustrate his theory of progressive sterilisation of potentially sporogenous tissue and does not need further mention here.

In cases where the potential mother-cells are selected in groups of two or more, it often happens that only one reaches maturity while the others suffer either an arrest in growth or divide into smaller cells. Either of these cases give rise to appearances which are likely to be interpreted in a different light. Sometimes we meet with three such abortive cells adjoining one side of the fully developed mother-cell and it strongly suggests the tetrad of an archesporial cell. This appearance is only deceptive, as the study of the early developmental history of the sporangium shows that they are not derivatives of a single cell, but are only groups of cells which are differentiated from a uniform mass. At other times, the abortive mother-cells which have divided to become smaller cells surround the fully developed mother-cell so as to be interpreted on cursory examination as a special layer of tapetum. But a critical study of the developmental history reveals their true nature. The tapetal cells as will be seen presently are formed much later in the history of the development of the sporangium.

All the aborted mother-cells finally divide into smaller cells until they reach the size of the cells of the sterile portion and are indistinguishable from them either in appearance or in behaviour. The fertile mother-cells continue to increase in size. They are at first polygonal in outline, but gradually assume a rounded or elliptical outline. By the time the mother-cells attain their maximum size, the sterile cells have practically ceased to multiply. Further changes leading to the differentiation of the sterile tissue into trabeculæ, tapetum and wall-cells start only after the mother-cells begin to divide.

While confirming Wilson Smith's observations on the origin, development and differentiation of the sterile tissue, further observations were made as to the formation of the cavities inside the sporangia. They arise by the disintegration of the cells surrounding the mature mother-cells. These cells very soon lose most of their contents, their nuclei only persisting for a time, while the

remaining cells of the sterile tissue possess a highly vacuolated protoplasm so that at this stage three different categories of cells are noticeable in the megasporangium: (1) The megaspore mother-cells in the early division stages marked by denser protoplasm; (2) The cells immediately surrounding the above with only the nuclei as their contents; (3) The rest of the tissue with cells having a nucleus and a highly vacuolated cytoplasm. (fig. 11). The latter give rise to the trabeculae and the tapetal tissue, whereas
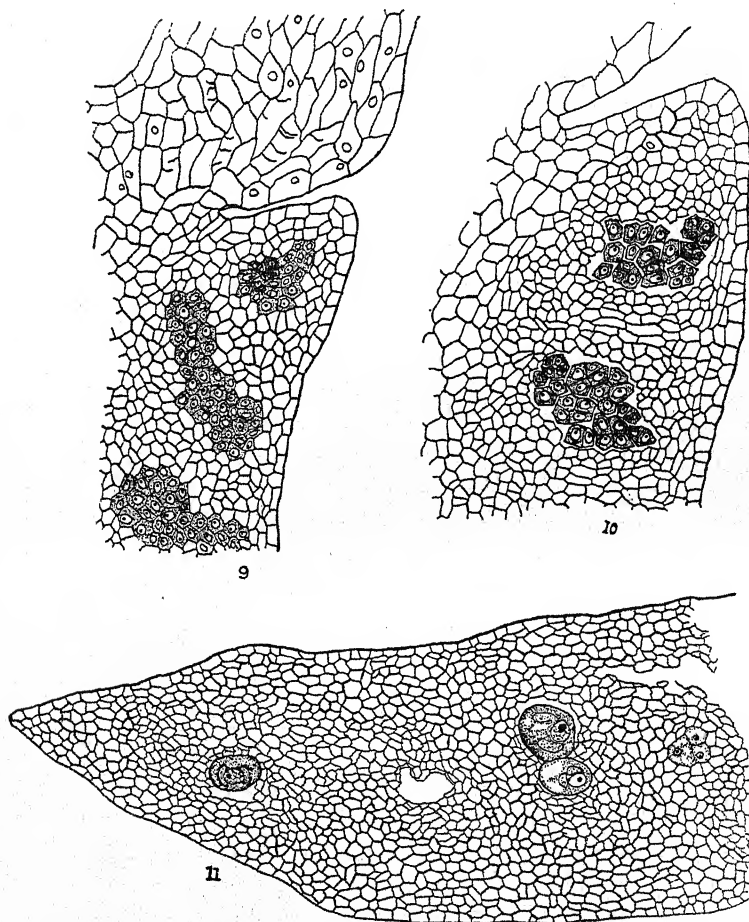
Fig. 9. A portion of a microsporangium showing the differentiation of groups of mother-cells. × 300. Fig. 10. The same; more advanced stage; the groups falling off into smaller ones just before division. Fig. 11. A portion of the megasporangium showing the degenerating sterile cells surrounding the mother-cells; Semi-diagrammatic. × 300.

the second category of cells disintegrate and are finally dissolved to form the cavities of the sporangium. After the degeneration of these cells the elongation of the trabecular cells takes place and the tapetal layers are differentiated.

The Microsporangium:—In the microsporangium, the course of development is more or less similar. The delimitation of the uniform mass of sporogenous tissue into fertile and sterile areas takes place relatively later, and groups of cells are selected to become the microspore mother-cells. (fig. 9). Here there is neither an arrest of growth nor a degeneration of the selected potential mother-cells, as seen in the megasporangium. All of them mature and undergo reduction division. Here also the multiplication of the cells of the sterile areas ceases by the time the mother-cells attain their full size, but the differentiation of the sterile tissue into trabeculae and tapetum takes place immediately so that one could recognise the latter even before the initiation of the divisions in the spore-mother-cells (fig. 10). There is no degeneration of the sterile tissue leading to the formation of cavities inside the sporangium as in the case of the megasporangium. Only a few cells lying close to the tapetum degenerate.

As to the details of development of the sterile tissue, the present species differs from *I. engelmanni* and *I. echinospora,* as described by Wilson Smith (36), in a few minor details. According to Wilson Smith, along with the elongation of the trabecular cells in the lateral direction, their nuclei also change their form, becoming spindle-shaped so as to resemble the plerome cells. In *I. coromandelina* the nuclei of the trabecular cells have not been observed to change their shape. Secondly, his observations show that the food materials, such as starch and oil, are not found in the cytoplasm of the trabecular cells. While discussing the functions of the trabeculae, Wilson Smith counts this fact as going against the suggested view that the trabeculae function as channels through which the nutrient material is supplied to the spores. He is of opinion that the food stuffs pass to the spores through the inner wall of the sporangium. The observations made here, however, reveal the presence of numerous starch grains, fat and oil globules in the cytoplasm of the trabecular cells. This would show that these cells function as channels for the supply of nutrient materials to the spores.

When the sporangium is fully ripe the sterile regions lose all their contents. The spores lie free in the cavities between the trabeculae and the sterile parts later on decay and the spores are

liberated. In the decay of the sterile regions the wall of the sporangium persists intact while the other parts, especially the portion attached to the leaf, break up into fragments. As such while the tubers are dug up for the collection of spores either very late in the season or just before the plant starts growth, one finds the spores freely falling away while the intact upper walls of the sporangia remain on the corm.

Occasionally, a few megaspores which do not reach maturity have been observed to develop in the microsporangium. Similar observations have been made by Wilson Smith (36) and Takamine (31).

The Megaspore Mother-cell.
The Reduction Divisions.

A fully developed megaspore mother-cell is ellipsoidal in outline. It has a dense, richly granular cytoplasm and a fairly big-sized nucleus. In the cytoplasm a number of fat globules are present which stain black with haematoxylin in the case of material killed in osmic acid fixatives and often obscure the other important details of the cell. The nucleus is situated a little towards one side of the cell and has a faintly staining reticulum and a prominent nucleolus which stains dark in haematoxylin, (fig. 12).

Heterotypic Prophase:—The first visible change initiating division in the mother-cells is the appearance in the cytoplasm of a body very close to the nucleus. This is clearly made out when it forms a deep crescent adjoining the nucleus. (fig. 13). It has a clear outline and appears to contain some dark staining granules of various sizes and other minute grains whose nature could not be made out with the methods of staining employed. Strasburger (30) and Tschistiakoff (32) called attention to the formation and division of this body in the megaspore mother-cells of *I. lacustris* and *I. Durieui.* As briefly referred to in the earlier part of the paper, Fitting (11) later gave a clearer account of the same. Observing them in the living condition, he describes this body as consisting of a coarsely granular plasm and starch grains and calls it in his description as a "starch clump". From the observations detailed below, it is found that this body is not merely a starch clump or starch body, though it often appears to contain a few starch grains. But its behaviour appears to be more of the nature of a centrosome and will be referred to as a "polar body" in this account.

As this "polar body" becomes conspicuous, the cytoplasm gets denser around it covering the whole of its convex surface. Very soon it shows a constriction in the middle which becomes deep, the cytoplasm at the same time getting sparse at this portion (fig. 14). Meanwhile the chromatin reticulum of the nucleus becomes a little more prominent. The next change is the division of the polar body into two at the constriction. The resulting halves move to the opposite sides of the nucleus and gradually increase in size. Several granules of irregular sizes are prominent at this stage within the bodies (fig. 16). Elongation now occurs in the two polar bodies at right angles to each other. At this time a few starch grains could be made out inside in addition to the irregular granules and other minute grains already referred. These starch grains could also be made out clearly in preparations stained with gentian violet. By now, the nucleus lies in the centre of the cell and its reticulum is well marked out. The nucleolus very often shows protruberances—a case of nucleolar budding observed commonly by other investigators (fig. 15).

A little later the nucleus is found to have again moved to one side of the cell. It now grows bigger in size with the growth of the mother-cell as a whole and its reticulum is gathered into a knot on one side of it, showing synizesis. This synizetic knot is so densely stained that its internal structure could not be made out (fig. 17). At this stage a second division of the polar bodies takes place. First the two bodies get connected by strands of cytoplasm. These strands are thick and anastomosing and are to be clearly distinguished from the spindle fibres which arise later. Material fixed with solutions of mercuric chloride shows these cytoplasmic strands very clearly (fig. 18). After the formation of these strands the bodies undergo constriction in the centre and each is divided into two. The cytoplasmic strands persist right through the process of meiosis.

With the second division of the polar bodies the open spireme stage of the nucleus is reached, the reticulum at this stage being much more prominent with several knots upon it. (fig. 20). Several stages of the second division of the polar bodies and the unravelling of the synizetic knot are met with in the preparations (figs. 18 & 19). This is the period of marked growth of the mother-cell which gradually becomes more or less spherical in outline. The cell-wall also becomes more prominent by the addition of a specially thin layer. The differentiation of the sterile

2

sporogenous tissue into trabeculae and tapetum occurs by this time and the megaspore mother-cells are seen lying free in the sporangial cavities.

The resulting four polar bodies sooner or later get roughly spherical and move away from the nucleus towards the periphery of the cell (figs. 21–24). They are always covered by a dense layer of cytoplasm and several anastomosing cytoplasmic strands are seen extending between them. The few starch grains inside these polar bodies show a further increase in number and it is at this stage that the grains could be well seen. They are not clearly made out in the slides treated with iron-alum haematoxylin but can however be easily seen in sections stained with saffranin and gentian violet (fig. 21). The starch grains do not stain with the same intensity as those present in the cells at the base of the leaf subtending the sporangium.

The amount of starch inside these bodies is variable; at no time are they filled with starch as to justify their denomination as mere "starch clumps". Usually a few light staining grains are present; more often there are none at all. This observation was further confirmed by iodine test on sections of living material collected during the season.

The megaspore mother-cell from this stage onwards shows little increase in size. The four polar bodies are finally arranged tetrahedrally near the wall of the mother-cell, each surrounded by a layer of cytoplasm which is denser towards the cell-wall. Henceforwards these polar bodies do not show any important changes being fairly constant both in shape and position.

Diakinesis:—Prominent changes follow in the nucleus. The reticulum is replaced by short thick chromosomes. The changes leading to the formation of the chromosomes are as follows: (1) a few local thickenings appear on the darkly stained spireme, which in course of time become conspicuous as deeply staining dark dots (fig. 22). (2) the chromatin is observed to be grouped into a few dark strands which are arranged more or less parallel to each other. (3) the reticulum is formed again with the threads now diverging rather widely and showing the peculiar twisting and interlacing that is commonly observed prior to the second contraction (fig. 23). (4) no definite "diffuse" stage or a regular second contraction was observed beyond the one shown in fig. 24, which may be taken to represent this stage. (5) the condensation of the spireme threads to smaller groups which finally shorten and thicken to form the bivalents (fig. 25).

Simultaneous with these changes, the nuclear outline takes a roughly triangular form in sectional view the angles of which point towards the polar bodies at the periphery of the cell. The spindle fibres now make their appearance. They are tetrapolar, each group of fibres proceeding from one polar body. These delicate fibres are clearly distinguished from the anastomosing cytoplasmic strands connecting the polar bodies and are well seen passing through the dense cytoplasmic sheath and are connected to the limiting membranes of these bodies.

Heterotypic Metaphase :—After the formation of the bivalents, the spindle fibres apply themselves to the nuclear membrane which gets dissolved at these places. Eventually the whole nuclear membrane disappears (fig. 26). The nucleolus which has shown no further visible change beyond the few buds given out at first also disappears at this stage (fig. 27). It has been, however, sometimes observed to persist for a short period after the dissolution of the nuclear membrane. The dissolution of the nuclear membrane and the disappearance of the nucleolus seem to take place gradually, as several intermediate stages are commonly met with in the preparations. Eventually the tetrapolar spindle becomes bipolar and the bivalents arrange themselves on the equatorial plate. Rarely the spindle fibres are bipolar without passing through the tetrapolar condition. The cytoplasmic strands connecting the polar bodies are now less prominent though persisting. The cytoplasm shows a slightly vacuolate condition at this stage.

It has been possible to count the number of chromosomes both at diakinesis and on the equatorial plate. Owing to the big size of the nucleus it is difficult to find all the bivalents in view in sections of less than 10 U in thickness. The counts in a large number of mother-cells show that the haploid number is 16. The same number has been made out by the counts in the microspore mother-cells also.

The metaphasic spindle (fig. 29) is of the ordinary type with one characteristic that it lies always between two of the polar bodies and the fibres are in close connection with the bodies themselves. Sometimes each of the remaining two polar bodies move towards the other two so that at each pole two of them are found very near each other.

Anaphase and Telophase :—The anaphase of the first division could not be observed in any great detail. Very few mother-cells were found in this stage and only the early (fig. 31) and the very late (figs. 32 & 33) conditions could be seen. The chromosomes

are unequal in size and they do not move towards the poles simultaneously. A few lag behind while the majority have reached the destination. Finally they collect at the poles very close to the polar bodies. The spindle fibres persist at late anaphase between two groups of chromosomes. (fig. 33).

The telophasic transformations follow quickly and with the appearance of a fresh nuclear membrane a temporary resting condition is reached. The spindle fibres disappear from the polar regions while becoming more prominent at the equatorial region. (fig. 34).

Interphase:—The period between the two divisions, though short, is characterised by a few important changes. The spindle fibres which were prominent at the equator during the late telophase are replaced by a cell-plate. This, however, is evanescent and disappears quickly. This evanescent cell-plate has been observed to be a peculiar feature of some reduction divisions. The daughter nuclei which were close to the polar bodies during the heterotypic telophase now move to a position between the two adjacent polar bodies (fig. 35). This resting period is very short and the second division follows almost immediately.

Homotypic division:—This is marked by the disappearance of the evanescent cell-plate and the quick prophasic changes that occur in the daughter nuclei. It was not possible to follow the various changes and the stage most commonly observed was when the bivalents were fully formed (fig. 36). During the following metaphase, the two spindles which are always at right angles to each other are free from one another (fig. 37)—a feature in contrast to that of the Lycopodiales and the Gymnosperms where sextuple spindles occur. Here again, each of the two spindles extend between two polar bodies. The chromosomes have the linear shape which they possessed during the anaphase of the first division. The changes that take place in the daughter nuclei are not always simultaneous; often one has reached the metaphase before the other has formed the bivalents. The only other stage observed of this division was the reorganised nuclei in the late telophase, one lying very close to each of the polar bodies. Now the cytoplasmic strands connecting the polar bodies become once again prominent.

Wilson Smith (36, p. 255) writes: "While the young megaspores have invariably a tetrahedral arrangement, occasionally a bilateral arrangement is found in which case the divisions so far observed are successive". He gives two figures in support of this

statement. The cells drawn in these figures (figs. 60 & 61, Pl. 18) cannot represent, in our opinion, the fully developed mother-cells which have undergone the reduction divisions, because they show vacuoles in their interior, are bounded on all sides by sterile cells and do not lie free in the cavities between the trabeculae. It has been pointed out that during the prophase of the first division the mother-cells get separated from the neighbouring sterile cells and come to lie freely in the cavities formed by the degeneration of the sterile cells. On the other hand, the mother-cells figured by Wilson Smith look more like degenerating and abortive ones. In the present study no case was met with where the division was successive. The interkinetic cell-plate was always evanescent and never formed a permanent cell-wall.

Cell division and *Spore formation* :—Cytokinesis results from the formation of a fine cell-plate between the four daughter nuclei along the equatorial plane (fig. 39). In the absence of the anaphase of the first division in our preparations, it was not possible to determine whether the plate was formed in connection with the nuclear spindle or independent of it. A little later this fine cell-plate splits along its whole length thus separating the four daughter cells. Very often, during fixing, the protoplasm of the daughter cells shrinks along this line and clearly brings into prominence the line of splitting of the plate (fig. 40). Each of the daughter cells secretes around itself a cell-wall and the cell-wall of the mother-cell is still intact enclosing the daughter cells (fig. 41).

A period of growth ensues, when several changes take place both inside the daughter cells and in their walls. The cytoplasm gets thinner with the formation of several big vacuoles. The nucleus remains at the base of the tetrahedral spore. It has now lost the staining capacity; the reticulum is very lightly stained, while the nucleolus is prominent. During the period of growth the nuclei are very irregular in shape but are ultimately rounded in the fully developed spores. The polar body one of which is persisting in each of the daughter cells shows a gradual diminution in size. Though it is prominent with the cytoplasmic covering during the formation of the daughter cells, it is not clearly made out in the enlarging spores enclosed in the mother cell-wall (fig. 42). In the fully developed spore it is totally absent

With the further growth, the cell-walls of each of the daughter cells increase in thickness and are finally differentiated

into three layers. The deposition of the wall along the line of partition between the daughter cells proceeds from the periphery to the interior and various stages of deposition of the wall, along this line of partition can be observed. Living material is more favourable for this observation. When the spores are more or less fully ripe the mother cell-wall is broken up and gets disorganised, and the spores come to lie freely in the sporangial cavities.

A fully developed megaspore is tetrahedral in shape with the wall differentiated into three layers. The outermost layer is marked with large tubercles. The cytoplasm shows a spongy structure and the nucleus is now found near the apical angle (apex) of the spore. (fig. 43).

Microspore Mother-cell

In spite of the rarity of the microsporangia referred to in the earlier part, we were fortunate enough to follow in detail the important phases of the reduction division. As the main course of events here is very similar to that in the megaspore mother-cells only a brief description will be given, pointing out a few interesting and peculiar features not found in the megaspore mother-cells. It has to be stated in the beginning that here also the polar body makes its appearance just before the inception of the division (fig. 45), going through the same changes as in the megaspore mother-cells and behaving as centrosome-like bodies at the poles of the spindle. The presence of this body in the microspore mother-cell has not been recorded by any of the previous workers.

The divisions start even when the mother-cells are in small groups retaining their polygonal outline. It is only when the division has progressed beyond synizesis that the individual cells fall apart from one another and get rounded. As noted by Wilson Smith (36, p. 249), hundreds of mother-cells in a microsporangium show more or less the same division stages. Separate "blocks", each showing a different stage of division, as noted by Ekstrand (10) were not observed in the present species.

The late prophasic changes, as the formation of the spindle fibres, the dissolution of the nuclear membrane and the disappearance of the nucleolus, have not been observed. But when the nuclear membrane is dissolved and disappears from view, the nuclear area is clearly recognisable from the cytoplasm and this condition persists through the division stages (see figs. 57–62). This is

something unusual, as it is generally the case that either the two portions are indistinguishable or are delimited by a special peri-nuclear mantle.

During metaphase (fig. 57), the spindle here also lies always between two of the polar bodies, the spindle fibres being connected with them. In hundreds of cells where the spindles were observed there was not even in one case where it occupied any other position.

In contrast to that of the megaspore mother-cells, the ana-phase of the first division could be studied in all its details. The movement of the chromosomes does not take place at the same time and they may be observed in several positions from the equator to the poles in a single cell. The two groups of chromo-somes are, however, distinct and the total number of each group is 16. (fig. 60).

Finally when all the chromosomes reach the poles near their respective polar body, they are grouped into a star-shaped mass (fig. 62). The nuclear membrane reappears in the very early telophase, the chromatin remaining still as a mass and not spread-ing itself into the whole of the nuclear area. (fig. 63). This is perhaps an unusual feature. We have already referred to the nuclear area being distinct even during the chromosome division. It would be a matter of interest to know if there is any correlation between these two features.

The daughter nuclei are as big as the nucleus of the mother-cell. The nucleolus reappears with the diffusion of the chromatin throughout the nuclear area (fig. 64). The spindle fibres form a fine plate at the equatorial region, which is either evanescent or persisting so that the division here is either simultaneous or suc-cessive.

During the second division, the spindles have been observed to be both parallel to one another (fig. 67) and at right angles to each other. In whatever position they may be, each spindle is free from the other and the two are never connected. The nuclei result-ing from the second division are arranged tetrahedrally (fig. 68) or bilaterally as the case may be. The polar bodies have not been followed throughout the stages of the second division, the prepara-tions made being not quite favourable for such observations. But they are nevertheless recognisable by the side of the nuclei after the second division.

The daughter cells soon get separated from each other. The wall of the microspore mother-cell disorganises very early so that

the spore-tetrads come to lie freely in the sporangial cavities at a very early stage (fig. 69). They remain in this condition for a fairly long interval. The spores, soon after their liberation from the mother-cell, possess a semi-lunar shape; their nuclei are very small in size when compared to that of the mother-cells. During this and the following stages the polar bodies are not present in the spores. A period of growth follows when they gradually attain a bean-shaped structure (fig. 71). The cytoplasm gets thinner and finely vacuolated; the cell-walls become differentiated into the usual three layers as in the case of the megaspore; the nucleus grows a little bigger in size though it never reaches the size in the mother-cell.

A fully developed microspore is bean-shaped and its wall differentiated into the three layers. The outermost wall is marked with small densely packed spines (fig. 72). Double spores are commonly met with. In the dry sporangium, the spores are powdery and reddish-brown in colour.

Discussion.

The "polar body":—While considering the nature of this body one has to bear in mind the following chief facts about them:

1. It is not seen in the early stages of the developing mother cells. It arises only just before the initiation of the prophase, in the cytoplasm of the mother-cells very close to the nucleus on one side of it.

2. It divides twice during the early prophasic changes of the nucleus.

3. The resulting four parts arrange themselves tetrahedrally on the periphery of the cell after which prominent nuclear changes follow.

4. The polar bodies are connected with one another by thick anastomosing cytoplasmic strands.

5. The contents of these bodies in the early stages are made up of irregular rods and other minute granules which give no reaction with iodine. But later starch grains, which fluctuate in quantity, appear.

6. The spindle fibres take their origin from these bodies and the nuclear spindle when differentiated always lies between two of these bodies. Hence the name polar body suggested.

7. After the conclusion of the division each daughter cell receives one of these bodies along with the nucleus. In the final stages of spore formation these bodies gradually diminish in size and ultimately disappear.

The occurrence of similar bodies has been recorded by previous workers in some other species of *Isoetes*. As has been referred to already, Fitting (11) found these bodies in the megaspore mother-cells of *I. Durieui* and *I. lacustris* and noted an essentially similar behaviour in them. He does not enter into a discussion as to the nature of these 'starch clumps' as he calls them, but draws the attention of the reader to the theoretical significance of the cytoplasm and its inclusions initiating the nuclear divisions in a few cases even among the Archegoniateae.

Marquette (19), while giving an account of mitosis in leaf cells of *I. lacustris* describes similar polar bodies in the cells emphasising on their fluctuating starch content. He calls them starch bodies throughout his description pointing out in the end, however, the possible objections to the name and suggesting the name ''polar structures''. After a brief discussion on the nature of these bodies he concludes ''It might be possibly assumed that in *Isoetes* we have a transition from a cell structure with well defined central bodies as found in some Algae and Fungi to a cell structure apparently without central bodies or anything corresponding to them as found in the spermatophytes''.

Ma (17), who studied the very young leaves and sporelings of *I. melanopoda,* found in the cells near the base of very young leaves the 'polar structures' described by Marquette in *I. lacustris.* By a comparative study of successively older cells of the leaf she endeavours to prove that numerous plastids in the older cells of the leaf are derived from the polar structures of the young leaves, though she does not observe the actual division of the structures and their formation into plastids. ·

It may be stated here that the bodies are met with in the dividing vegetative cells of *I. coromandelina* also. Our few observations on mitosis, both in the cells of the intercalary meristem of the leaf and that of the young sporogenous tissue, agree with those of Marquette as to the structure and behaviour of these polar bodies. The division of these bodies to form the plastids of older cells, as described by Ma for *I. melanopoda,* was not observed to take place.

Bodies of similar nature have also been found in the case of a few Bryophytes as *Anthoceros* and certain Mosses. Strasburger

(30) was the first to describe the two repeated divisions of a body in the spore mother-cells of *Anthoceros*. Since his work, numerous others have also observed it in other species of *Anthoceros*. Campbell (8, p. 141, fig. 73 A-D.) followed the developmental history of the spores in most of its details in the fresh material of *A. fusiformis*. He describes a roundish body lying very close to the nucleus, of granular consistence and yellowish green colour, containing a number of granules some of which are starch. Davis (9) worked in detail on the spore-mother-cells of *A. laevis* and his observations are mainly from permanent preparations. He was unable to find in the young cells of the sporogenous layer any body of the nature of plastid or chloroplast. According to him the body makes its appearance in the mature spore-mother-cells and is made up of starch grains, which stain sharply a beautiful purple with gentian violet, embedded in protoplasmic strands and fibrils. This chloroplast as he calls it behaves in a manner similar to the polar body in *Isoetes* here described. Here also the four bodies formed as result of two repeated divisions are connected to one another by coarse cytoplasmic strands; the nuclear spindle is at first tetrapolar taking its origin from the bodies; later a bipolar spindle is formed which always lies between two of these bodies.

In *Polytrichum*, Allen (1) found each pole of the spindle occupied by a flat polar plate which behaves as a division centre though not as an aster. This body persists in the resting condition, divides in the prophases of the divisions in the spore-mother-cells to form polar plates from which protoplasmic fibrillae grow towards the nucleus and finally form the spindle. In the second division the polar plate is represented by a group of granules which divide into two similar groups as before. In spermatogenesis this body has been observed to function as the blepharoplast.

Sapehin, in a series of articles (23, 24, 25, and 26), records his observations on the condition of plastids and their individuality in the sporogenous cells of Mosses (*Funaria, Polytrichum, Bryum,* and *Mnium*), *Selaginella,* and *Isoetes.* His main contention is that they are continuous throughout the life-cycle of a species and is convinced that the bodies in all the plants observed by him should be included in the category of plastids. He is also positive that plastids are not formed by chondriosomes which he thinks are quite independent cytoplasmic structures found side by side with the plastids.

McAllister's studies of *Anthoceros* and *Notothylos* (20) with regard to the structure of plastids in the gametophytic and sporogenous cells make him believe in the continuity of plastids throughout the life-cycle of the species studied by him. Recognising the presence of plastids in the young sporogenous cells and elater cells, his conclusions as to the nature of the bodies in the spore-mother-cells are: "Although the exact identity of the plastids in the developing spores and in the other reproductive cells is somewhat uncertain, there seems to be little doubt that these vacuolate areas in the spore-mother-cells of *Notothylos* and *Anthoceros* are plastids. They have arisen by the transformation of the structures which are undoubtedly chloroplasts and they occupy the same position as those structures occupied. The formation of starch bodies within these areas even though they are very unlike the conventional plastids is evidence of their plastid nature."

Ma's observations (18) on the sporogenous cells of certain Mosses (*Atrichum undulatum, Bryum argentinum, Funaria americana,* and *Physcomitrium turbinatum*) lead her to the conclusions of Sapehin and McAllister as to the continuity of plastids. But she has not followed closely the history of the body in the sporogenous cells of any one single species but makes only general observations on preparations which happen to show a few stages.

On the other hand, Senjaninova (28) who studied the plastids and chondriosomes of the sporogenous layer in *Catherina* and *Physcomitrium*, believes that the plastids disappear from the archesporial cells at about the time of differentiation of these cells. She describes numerous chondriosomes of various shapes as occurring in the archesporial cells. They, altogether with some of the cytoplasm, aggregate to form an irregular saucer-shaped structure curving about the nucleus. Senjaninova regards this structure as a complex plastid in which the elongated starch grains may be seen. She believes that these bodies undergo segmentation to form the plastids of sporogenous cells and spores.

The latest work on such bodies in Mosses is that of Weier (33) who has made some observations on the cytoplasmic bodies during sporogenesis and spermatogenesis in *Polytrichum commune*. A study of the plastid during sporogenesis after fixation by mitochondrial, osmium and silver techniques as well as the vital studies show that they have striking resemblances to the golgi bodies as found in the insects. She draws up a comparison between the

similarities in structure and behaviour of plastid and golgi bodies before and after meiosis in *Polytrichum commune* and *Euschistus* (Bowen 1920) and feels that there must exist some relation between the two *.

Sharp (29, p. 109) seems to be inclined to the view that the bodies under discussion are plastids, which remain as morphological individuals throughout the whole life-cycle of a species, assuming several shapes and sizes during certain stages of the life-cycle and multiplying exclusively by division. In these, the plastids possess an individuality comparable to the nuclei from which they differ conspicuously however in undergoing no fusion at the time of syngamy. But he does not commit himself to a definite conclusion as he feels that the problem of the origin of plastids is greatly complicated. The pole plates of *Polytrichum* as described by Allen. (1) are supposed by him to possess a problematical relation to the centrosomes. (l.c. p. 298).

Wilson (35, p. 201) regards the true nature of the "starch bodies" of *Isoetes* described by Marquette and the pole plates of *Polytrichum* described by Allen as still uncertain, but suggests that they might be taken to represent possible intermediate condition between the amphiastral and anastral divisions in plants.

It would be seen from the review of the work of the above mentioned authors that the observations of no two authors are similar and that each has been inclined to interpret the body in question as either a plastid or a centrosphere-like body. The observations here made in *Isoetes coromandelina* favour the view that the "polar bodies" are centrosomal in behaviour. They make their appearance in the cytoplasm just before the commencement of the division of the nucleus of the spore-mother-cells. It is not possible to observe the same body in the young sporogenous cells as it is absent in them at this stage. During the process of nuclear division they have a close association with the nucleus e.g. their invariable polar position and the formation of the achromatic figure in relation to these bodies. (See figs. 26 & 27). In the case of the spore-mother-cells of *Isoetes* it cannot be argued that these bodies come to occupy the polar positions by mere accident or chance. The spore-mother-cells are big enough to allow a free movement of these bodies in the cytoplasm and their position is

---

* Refer also to Weier, E. T., "The structure of the Bryophyte plastid with reference to the golgi apparatus" Amer. Journ. of Botany, Vol. 19, 1932 659–672.

not circumscribed and determined by the location of large masses of cytoplasm.   One cannot escape the view that there is a definite correlation between these bodies and the nuclear division, and that they perform the functions of a centrosome acting as division centres (figs. 29, 57 & 37).

The presence of a few grains of starch, which is essentially a plastid product, is the only evidence as to their plastid nature. But the other characters as to their centrosomal behaviour are too many to be easily ignored.   If we are to choose between the two views, whether the bodies are mere plastids or centrosphere-like bodies, we find that there is more evidence in support of the latter.   Further work as to the exact origin of the bodies in the mother-cells and their subsequent behaviour during spermatogenesis and oogenesis will be of much interest, especially in view of the fact that Belajeff (2, p. 795 and fig. 18) has noted two biscuit-shaped bodies at the poles of the nucleus before the liberation of sperms in *I. malinverniana* and Takamine (31, p. 186 fig. 2) observed bodies plastid-like in appearance in the egg cells of the fully developed archegonium of *Isoetes japonica*.

Summary.

1. The microsporangia are found in a very few plants.   Most of the plants are megasporangiate.   In those plants where the microsporangia occur, they are found among the outer sporophylls and not in the inner rosette of leaves.

2. The velum, which is present in this species as a rudimentary structure, arises from the tissue between the ligule and the sporangium.   Its origin and development show that it is an independent vegetative structure and not a sterilised portion of the sporangium.

3. A connection between the tracheids of the velum and the leaf-trace bundle is established.   This fact suggests that the velum is a fundamental part of the leaf, presumably of greater antiquity than the ligule.

4. The sporangium takes its origin from a superficial group of cells.

5. The early development of the two kinds of sporangia is similar up to certain stage when the differentiation into sterile and fertile regions takes place in them.

6. The spore-mother-cells in both the kinds of sporangia are selected out of a uniform mass of cells and there is no evidence of a hypodermal archesporium at any stage of development.

7. In the megasporangium all the megaspore mother-cells, though potentially sporogenous, do not reach maturity; only a few of them finally mature and undergo the reduction division to form the spores. The unsuccessful ones divide to form smaller cells which share the same fate as the sterile tissue.

8. In the microsporangium the changes occur relatively later than in the megasporangium. Here groups of cells are marked out to form the mother-cells most of which are capable of producing ripe spores.

9. The sterile tissue becomes transformed into trabeculae, tapetum and wall cells. In the megasporangium the tissue immediately surrounding the fertile mother-cells disintegrate and contribute to the formation of the cavities in which the mother-cells lie freely. In the microsporangium only a very few cells adjoining the tapetum degenerate.

10. The different phases of the reduction division are described in detail for both the mega- and microspore-mother-cells.

11. Preliminary to the reduction division of the nucleus a "polar body" appears in the cytoplasm of the mother-cells very close to the nucleus. This divides twice during the prophases of the first division and the four resulting parts arrange themselves tetrahedrally on the periphery of the cell, and are connected to one another by cytoplasmic strands. Later the nuclear spindle arises between these polar bodies. After the nuclear division they become less prominent and disappear in the fully developed spores.

12. The nature of this body is discussed and its centrosomal behaviour is emphasised.

13. The counts of chromosomes in the diakinesis and heterotypic metaphase of both kinds of spore-mother-cells and in the anaphase of the first division in the microspore mother-cells show that the haploid number for the species is 16.

Our thanks are due to Dr. M. A. Sampathkumaran, M.A., Ph.D., for many helpful suggestions and criticism.

Botanical Laboratories,
  Madras University,
    South India.

Literature Cited.

1. ALLEN, C. E.—Cell structure, growth and division in the antheridia of *Polytrichum*. Arch. f. Zellf. 8; 121–128; 1912.

2. Belajeff, W.—Antheridien und Spermatozooiden der heterosporen Lycopodiacean. Bot. Zeit. 43, 794–800; 1885.

3. Botanical Survey of India—Records. Vol. III. p. 327.

4. Bower, F. O.—Studies in the morphology of the spore-producing members : Equisetineae and Lycopodineae. Phil. Trans. Roy. Soc. Ser. B. 189, 35–81; 1897.

5.   „   The Origin of a Land Flora. London. 1908.

6. Bowen, R. H.—Studies on insect spermatogenesis, I. The history of the cytoplasmic components of the sperm in Hemiptera (*Euschistus*). Biol. Bull. 39, 316–360; 1920.

7. Braun, A.—Uber die Isoetes-arten der Insel Sardinien, nebst allgemeinen Bemerkungen uber die Gattung *Isoetes.* Monatsber. d. kgl. preuss. Akadamie d. Wiss. zu Berlin. 1863.

8. Campbell, D. H.—Mosses and Ferns. 2nd Edn. New York. 1918.

9. Davis, B. M.—The spore-mother-cells of *Anthoceros.* Bot. Gaz. 28. p. 89–109. 1900.

10. Ekstrand, H.—Uber die Mikrosporenbildung von *Isoetes echinospora* Dur. Svensk. Bot. Tidskr. Bd, 14, 312–318, 1920.

11. Fitting, H.—Bau und Entwicklungsgeschichte der Makrosporen von *Isoetes* und *Selaginella* usw. Bot. Zeit. 58, p. 107–165, 1900.

12. Goebel, K.—Beitrage zur vergleichenden Entwicklungsgeschichte der sporangien. Bot. Ziet. 38 and 39. 1880–81.

13. Gluck, H.—Wasser-und Swampflanzen., 4. p. 22–, 1924.

14. Hegelmaier, F.— Zur Kenntnis einiger Lycopodinen. Bot. Zeit. 32. 1874.

15. Hofmeister, W.—The Higher Cryptogamia. Roy. Soc. London. 1862.

16. Kienitz-Zerloff, F.—Ueber Wachsthum und Zellteilung in der Entwickelung des Embryos von *Isoetes lacustris.* Bot. Zeit. 39. 761–770. 1881.

17. Ma, R. M.—The chloroplasts of *Isoetes melanopoda.* Amer. Journ. Bot. 15, 277–284. 1928.

18. MA, R. M.—Starch deposition in the sporogenous cells of certain Mosses. Bull. Torrey Bot. Club. 51, 521–532, 1930.

19. MARQUETTE, W.—Manifestations of polarity in the plant cells which are apparently without centrosomes. Beih. Bot. Centralb. Bd. 21. Abt. 1. 281–303. 1907.

20. McALLISTER, F.—The pyrenoids of *Anthoceros* and *Notothylos* with special reference to their presence in spore-mother-cells. Amer. Journ. Bot. 14. 246–257. 1927.

21. MILDE, J.—Filices Europeae Atlantidis Asia Minoris et Siberiae. Leipzig. 1867.

22. PFEIFFER, N.—Monograph on the Isoetaceae. Ann. Miss. Bot. Gard. Vol. 9. No. 2. 1922.

23. SAPEHIN, A. A.—Ueber das Verhalten der Plastiden im sporogen Gewebe. Ber. der Deuts. Bot. Gessel. Bd. 29, 491–496, 1913.

24. „ Investigations on the Individuality of the Plastids. Odessa (Russian). 1913. (Quoted by Sharp).

25. „ Untersuchungen uber die individualität der plastiden. Ber der Deuts. Bot. Gessel. Bd. 31, 14–16 and 321–323.

26. „ Individuality of Plastids. Arch. f. Zellf. 13, 319–398, 1915.

27. SCOTT, D. H. and HILL, T. G.—Structure of *Isoetes hystrix*. Ann. Bot. 14, 413–454, 1900.

28. SENJANINOVA, M.—Origin of the Plastids during sporogenesis in Mosses. Zeitschr. Wiss. Biol. Abt. B. 6, 464–492, 1927.

29. SHARP, L. W.—Introduction to Cytology. 2nd Edn. 1924.

30. STRASBURGER, E.—Zellbildung und Zellteilung. 3te Aufl. Jena. 1880.

31. TAKAMINE, N.—Some observations in the life-history of *Isoetes*, Bot. Mag. Tokyo. Vol. 35. No. 418. 1921.

32. TSCHSTIAKOFF, J.—Notice preliminaire sur l'historie du developpement des sporanges et des spores de l'Isoetes Durieui Bory. Nuovo Giorn. bot. ital. 5, 1873.

33. WEIER, E. T.—A study of the moss plastid after fixation by mitochondrial, osmium and silver techniques. I. The plastid during sporogenesis in Polytrichum commune. La Cellule. Vol. 40. 261–290, 1931.

34. WEST, C. and TAKEDA, H.—On *Isoetes japonica*. Trans. Linn. Soc. ser. B. Botany. Vol. 8. pt. 8. 1915.

35. WILSON, E. B.—Cell in Development and Heredity. 3rd Edn. 1928.

36. WILSON SMITH, R. The sporangia and the sporophylls of *Isoetes*. Bot. Gaz. Vol. 29. 225–258; 323–346, 1900.

Explanation of Plates.

All preparations were studied under a Zeiss oil immersion apochromatic objective × 90. The figures were drawn with the aid of an Abbe's Camera Lucida and reduced to two-thirds the original size.

PLATE I.

Megaspore mother-cells in division stages.

Fig. 12. A fully developed megaspore mother-cell. × 630.

Fig. 13. The appearance of the polar body in the cytoplasm. × 630.

Fig. 14. Constriction and first division of the polar body. × 630.

Fig. 15. The two polar bodies opposite to each other; also nucleolar budding.

Fig. 16. Polar bodies showing the irregular granules inside them. Stained in picro carmine. × 630.

Fig. 17. Synizesis; cytoplasmic strands connecting the polar bodies prominent; killed in mercury chloride solution. × 630.

Fig. 18 and 19. Post-synaptic spireme; second division of the polar bodies. × 630.

Fig. 20. Open spireme stage of the nucleus. × 630.

Fig. 21. Four polar bodies with prominent starch grains; stained in saffranin and gentian violet. × 630.

Fig. 22. The reticulum showing local. thickenings. × 630.

PLATE II.

Megaspore mother-cells in division stages.

Fig. 23. Strepsinema. × 630.

Fig. 24. Second contraction. × 630.

3

Fig. 25. Diakinesis. × 630.

Figs. 26 and 27. Dissolution of the nuclear membrane and the disappearance of the nucleolus. Tetrapolar spindle formation. × 630.

Fig. 28. Equatorial plate. × 630.

Fig. 29. Metaphasic spindle. × 630.

Figs. 30–33. Heterotypic anaphase. × 630.

Fig. 34. Interphase; with the evanescent cell-plate. × 630.

PLATE III.

Fig. 35. Megaspore mother-cell after the first division with the resting daughter nuclei. × 630.

Fig. 36. The same; prophase of the second division. × 630.

Fig. 37. The same; second division. × 630.

Fig. 38. The same; showing the early anaphase of the second division. × 630.

Fig. 39. The same showing the formation of cell-plates at the equatorial region. × 630.

Fig. 40. Separating daughter spores. × 630.

Figs. 41 and 42. The developing spore tetrads. × 630.

Fig. 43. A fully developed megaspore. × 300.

Fig. 44. A small group of microspore mother-cells just before nuclear division. × 1250.

Fig. 45. A microspore mother-cell; the appearance of the polar body. × 1250.

Fig. 46. The same; first division of the polar body. × 1250.

Fig. 47. The same; two polar bodies moving away from one another. × 1250.

PLATE IV.

Microspore mother-cells in division stages.

Figs. 48 and 49. Two polar bodies opposite to one another. × 1250.

Figs. 50 and 51. Synapsis and second division of the polar body. × 1250.

Figs. 52 and 53. Post synaptic spireme. × 1250.

Fig. 54. Open spireme stage of the nucleus. × 1250.

Fig. 55. Segmentation of the spireme to form the bivalents. × 1250.

Fig. 56. Diakinesis. × 1250.

Fig. 57. Metaphase. × 1250.

Figs. 58–62. Anaphase of the first division. × 1250.

PLATE V.

Fig. 63. Microspore mother-cell; Telophase of the first division. × 1250.

Fig. 64. Interphase. × 1250.

Fig. 65. The same; prophase of the second division. × 1250.

Figs. 66 and 67. The same; homeotypic metaphase. × 1250.

Fig. 68. The same; cell-plate formation. × 1250.

Fig. 69. Young microspores soon after their liberation from the mother cell-wall. × 1250.

Figs. 70 and 71. The developing microspores. × 630.

Fig. 72. A fully developed microspore. × 300.
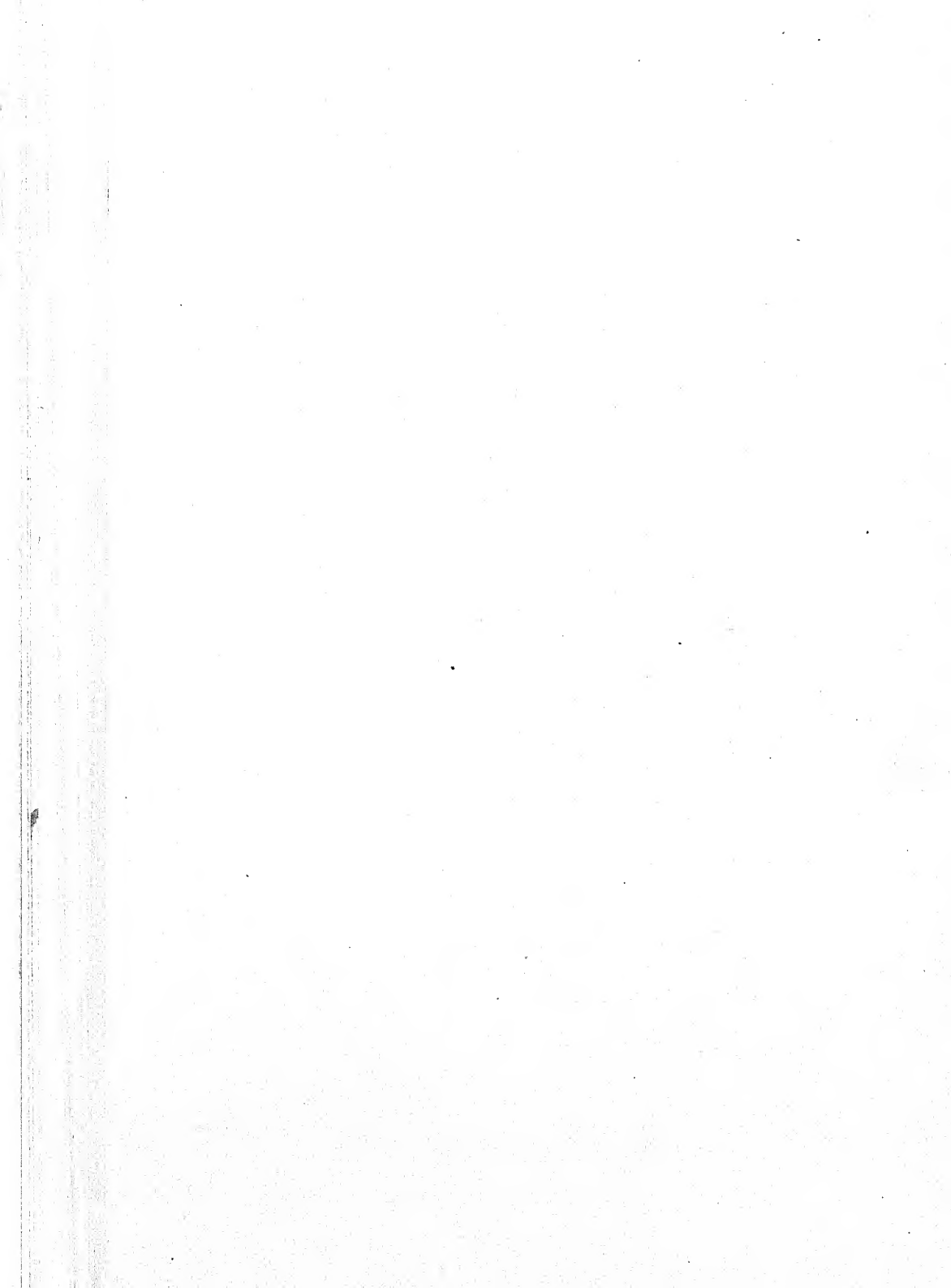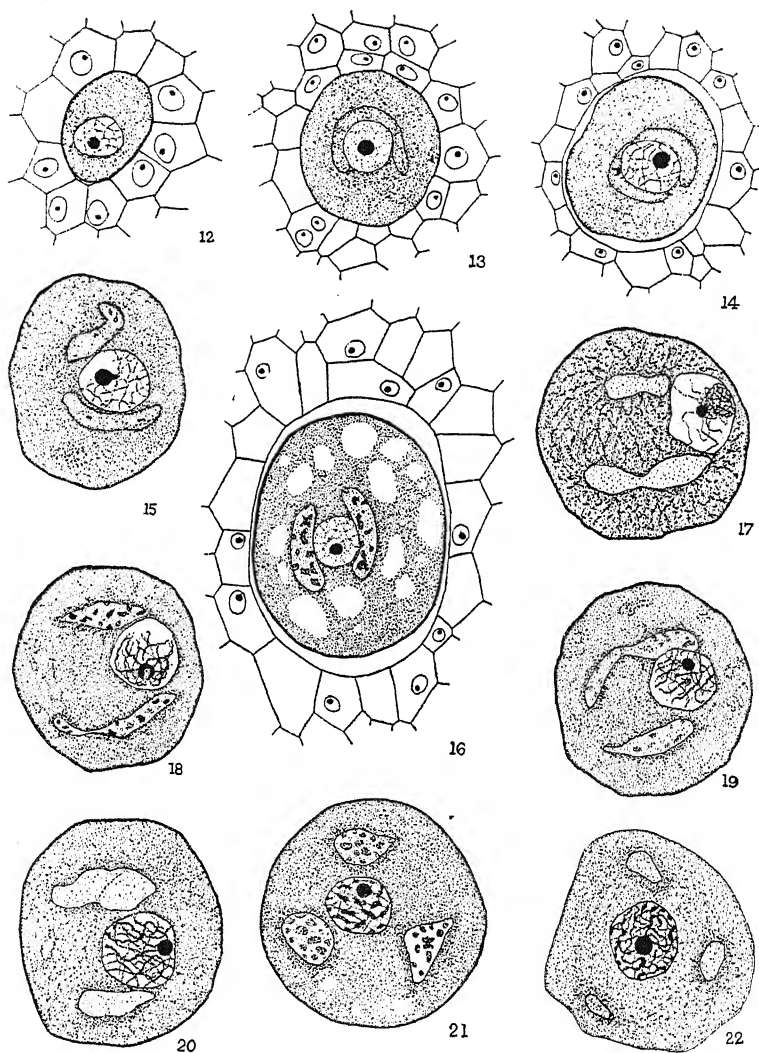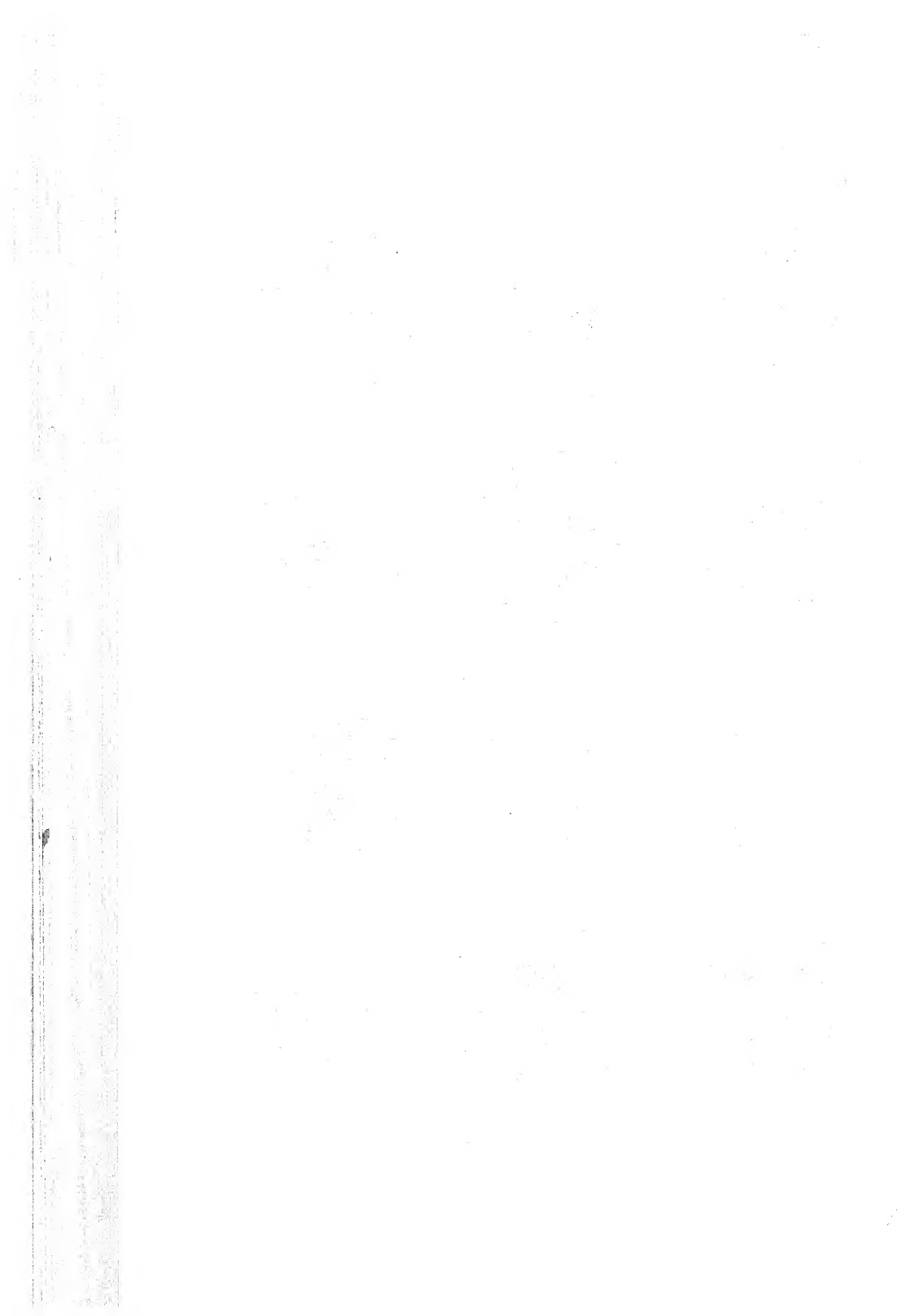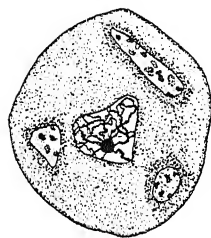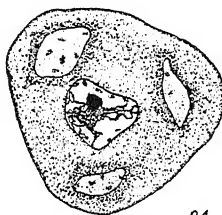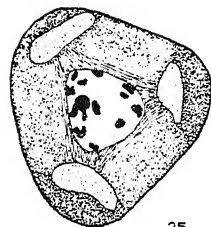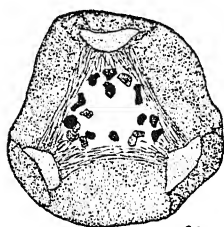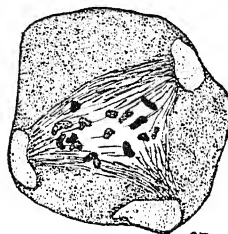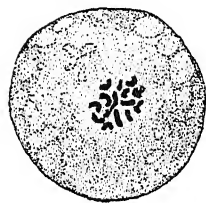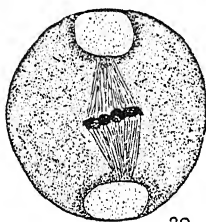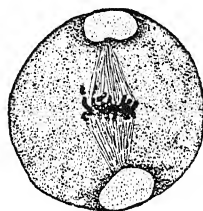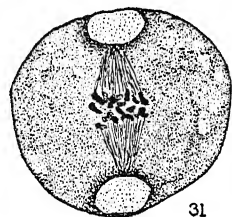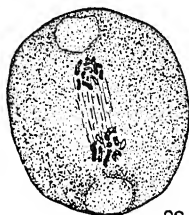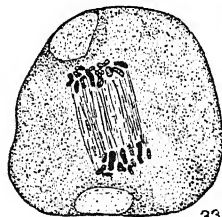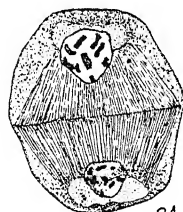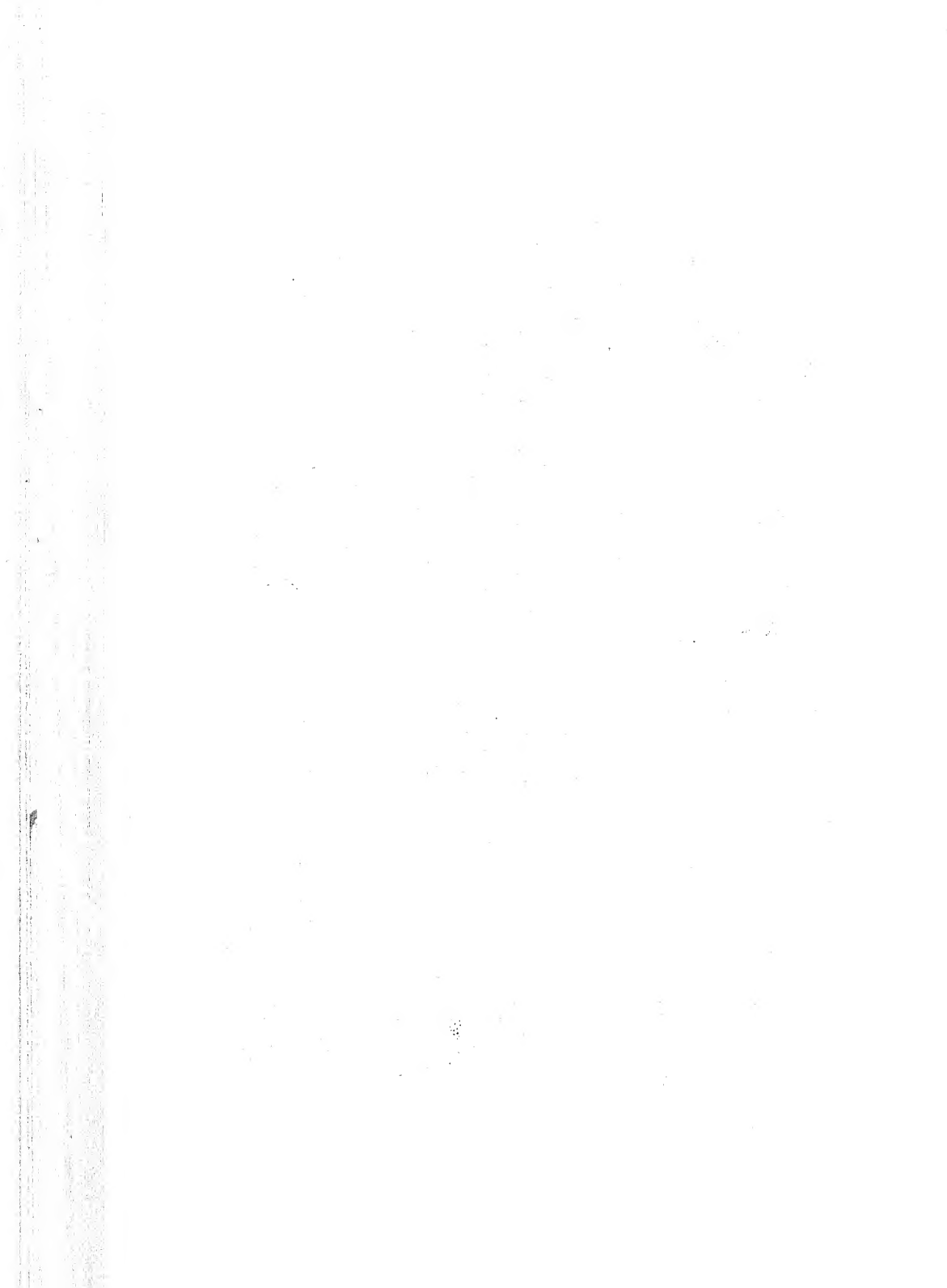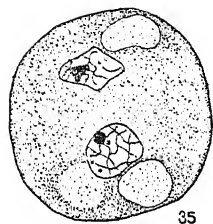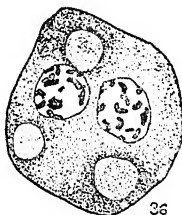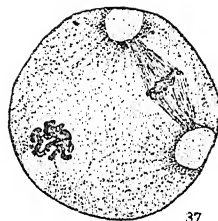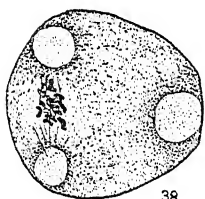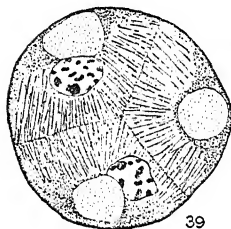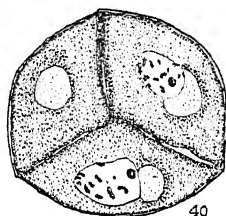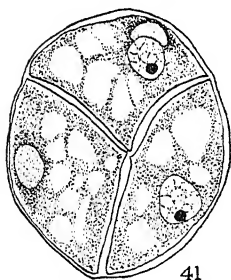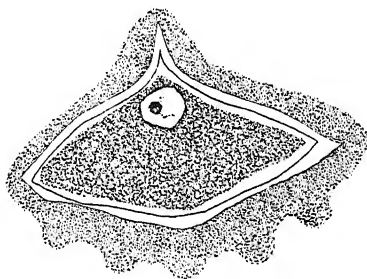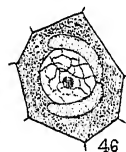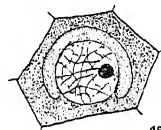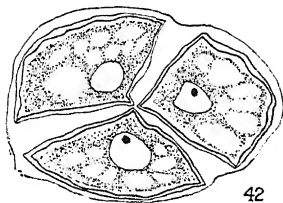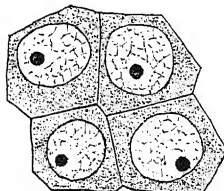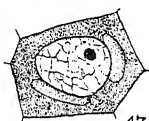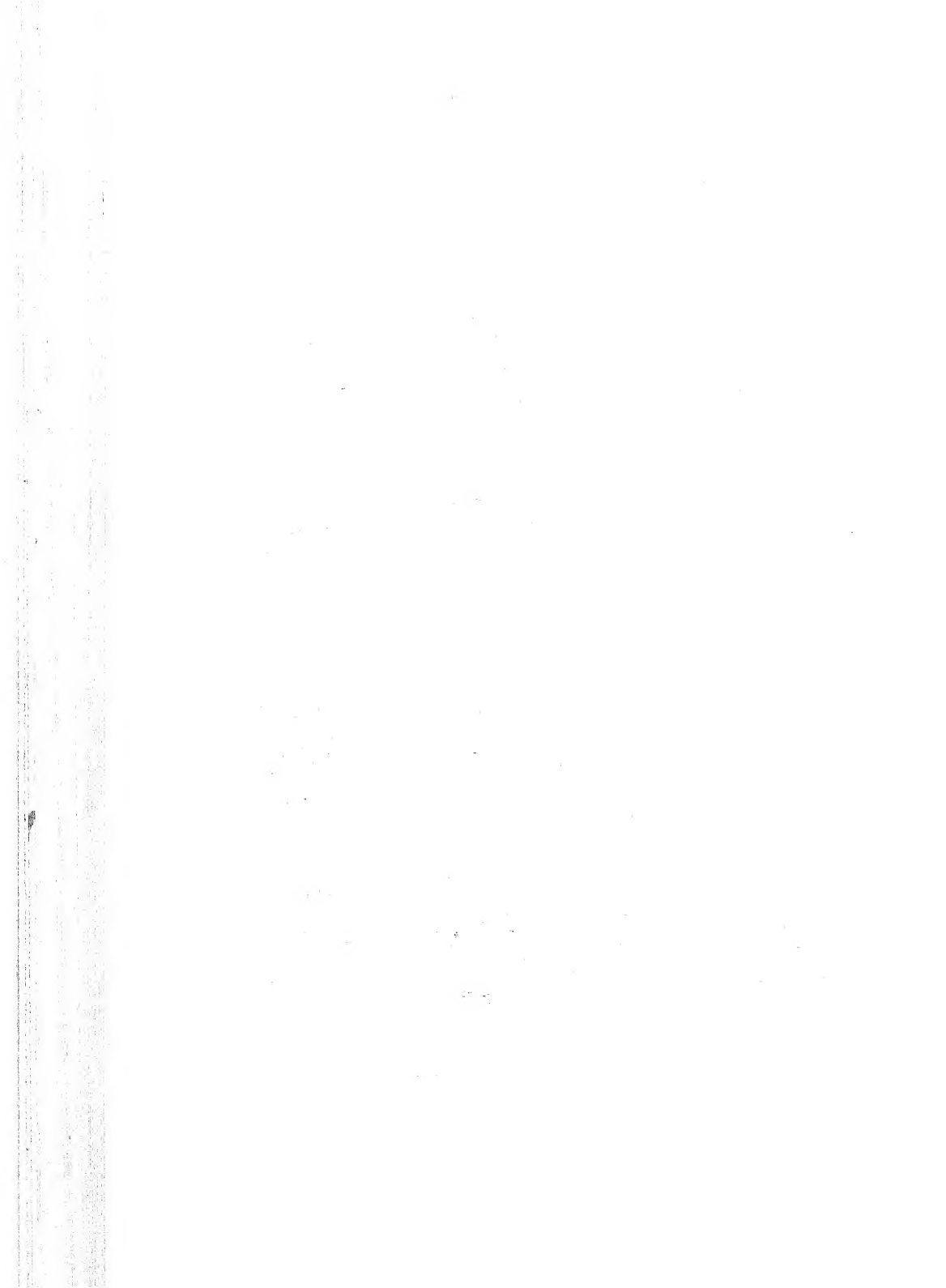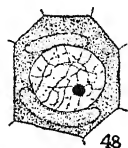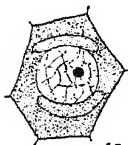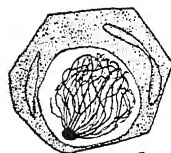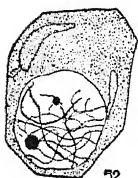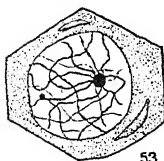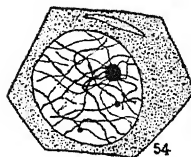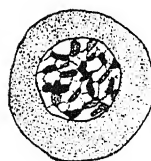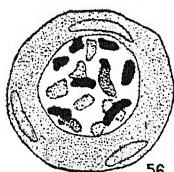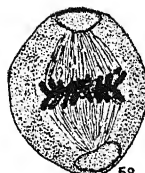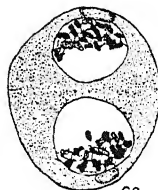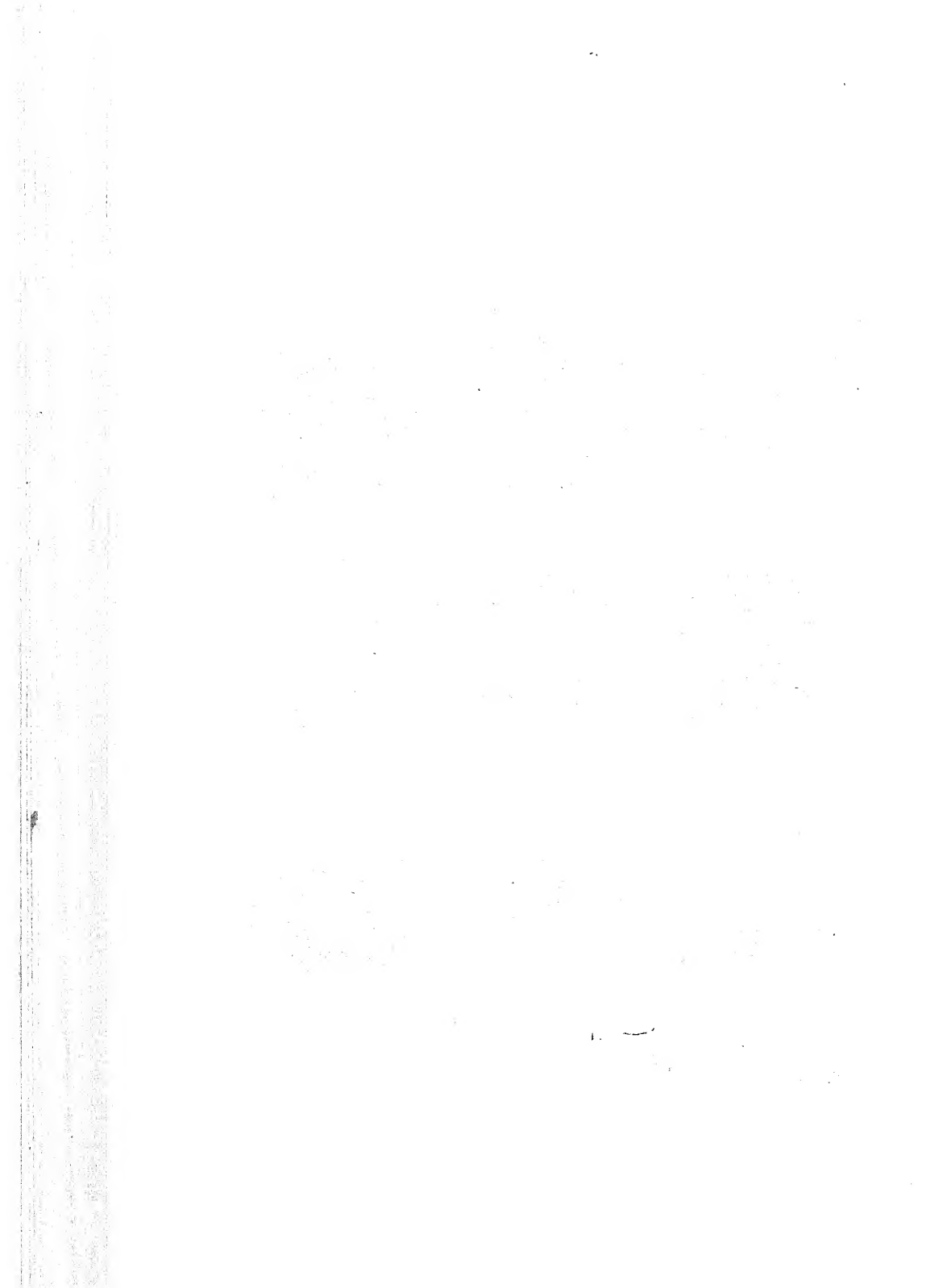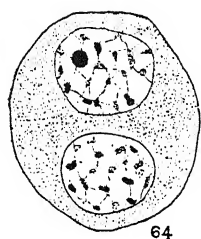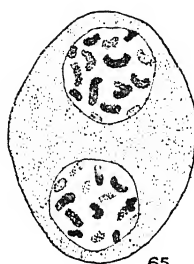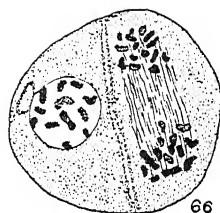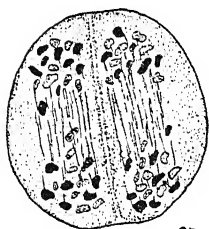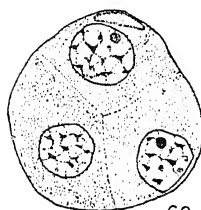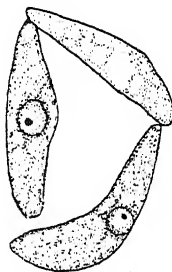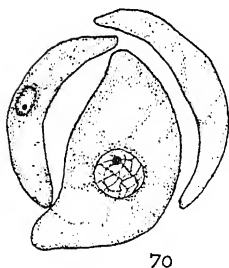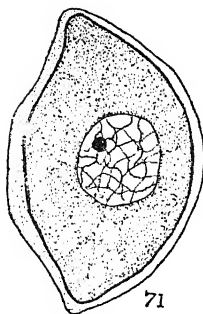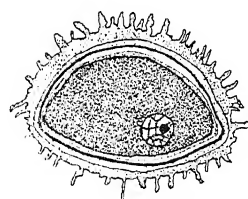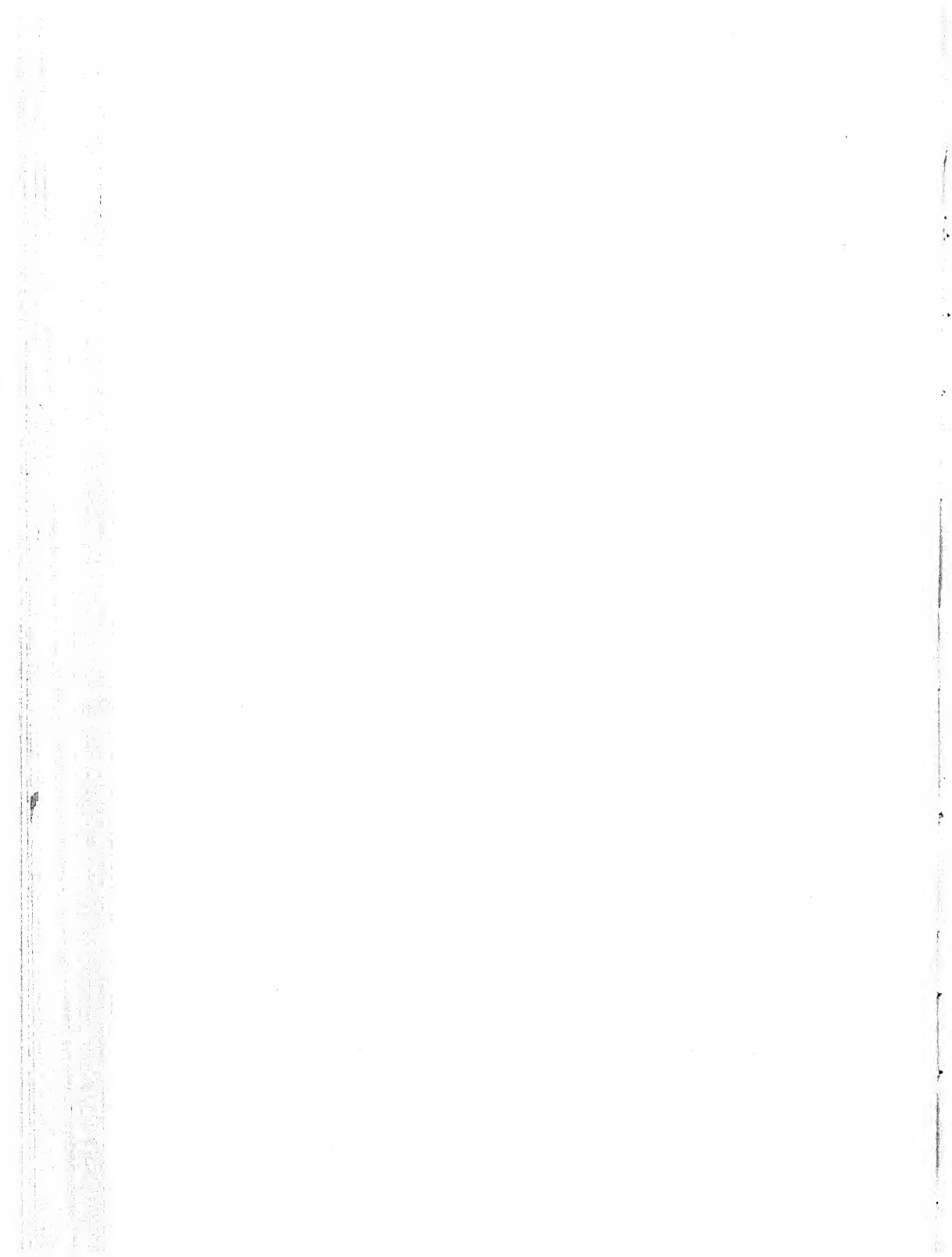
THE 'ROOT-THORN' OF *BRIDELIA PUBESCENS*, Kurz

BY

P. Parija and P. Misra,

Ravenshaw College, Cuttack.

Introduction.

The occurrence of 'root-thorn' is very rare indeed; the only recorded cases, as far as known, are those of *Acanthorhiza* (1, 2, 3, 4) and *Iriartea* (1, 2, 3) (Palmae), *Dioscorea prehensilis* (Dioscoreaceae) (1); *Moraea* (Iridaceae) (1) and *Myrmecodia* (Rubiaceae) (1); the last named being the only dicotyledonous genus. No mention, however, has been made in the available literature regarding the 'root-thorn' in *Bridelia pubescens* Kurz, belonging to the family Euphorbiaceae.

The observations presented in the succeeding pages were made on a small tree of *B. pubescens* growing in the garden attached to the botanical laboratory. A number of thorns were found growing at right angles to the axis on the main trunk as well as at the bases of the branches (Pl. I, fig. 1). Superficially they looked like stem-thorns grown from adventitious buds. Only during the rains they make their appearance and grow till about November and then turn into thorns. A splendid case of an old thorn producing many new branch-thorns at its tip in all directions at right angles, had also been noticed. In young thorns, the presence of a root-cap-like structure at their tips led us to suspect that they might be roots in origin and it was, therefore, thought desirable to undertake the investigation.

Anatomical Study

(i) *General:*

Longitudinal section of a young 'root-thorn' (Pl. I, fig. 4) shows a many-layered root-cap at the apex, behind it a many-layered calyptrogen which continues as the many-layered dermal tissue of the older region; next to it the periblem and last of all the central cylinder of plerome.

(1) Goebel, K.—Organography of plants, II, p. 288, 1905.

(2) Solereder, H. & Meyer, F. J.—Systematische Anatomie der Monokotyledonen, III, p. 81, 1928.

(3) Willis, J. C.—A dictionary of flowering plants and ferns pp. 10, 344 and 648, 1925.

(4) Small, J.—A text-book of botany, p. 110, 1929.

Most of the cells of the dermal tissue and of the cortex are packed with so much of coloured granular matter that it is very difficult to demarcate the two regions sharply in a longitudinal section. This granular matter appears greenish yellow in the young stage and does not change colour on being treated with iron salts. The contents have not been identified but they take stains very easily. Probably these cells are laticiferous. The cells of the cortex have slightly thickened walls and the thickening is of pitted type.

(ii) *Primary structure:*

In the stele of 'root-thorns,' the bundles are radial. It is polyarch, consisting of many (fourteen to twenty) exarch xylem bundles alternating with as many phloem bundles (Pl. II, fig. 4), while in the subterranean roots the number of xylem bundles is four to six (Pl. II, fig. 2). At a glance the transverse section of the 'root-thorn' looks like that of a monocotyledonous root; on the other hand the under-ground root conforms to the dicotyledonous type. So the 'root-thorn' is quite unlike the subterranean root.

The endodermis is of a single layer, all the cells of which are filled with a yellowish green pigment. The cells of the cortex are fairly thin-walled with simple pits on their radial walls, as these pits are visible only in longitudinal sections. Several cells of the cortex contain the characteristic pigment noted above. The outermost cells of the cortex form a distinct layer which delimits the cortical tissue from the many-layered dermal tissue and appears like an exodermis (Pl. II, fig. 5). Most of the cells of this layer contain the characteristic pigment already referred to.

The dermal tissue is four to five cells in thickness (Pl. II, fig. 5), looking like the velamen of the orchid root, but in this case the cells are living and have highly granular contents which are sometimes greenish yellow in colour. The cell-walls are thick but no pits or other patterns are seen. In the young 'root-thorns', excepting the outermost layer, all the cells of the dermal tissue are radially elongated. The outermost cells peel off as the age advances.

(iii) *Secondary structure:*

In the stele of the 'root-thorn', the formation of the cambium is just like that in ordinary dicotyledonous root (Pl. II, figs. 3, 6). The phellogen is formed either in the outermost cells of the cortex or in the cells just outside this layer (Pl. II, fig. 7). In the subterranean root the phellogen forms as in ordinary dicotyledon-

ous roots (Pl. II, fig. 1), i.e. in the pericycle; the endodermis, primary cortex and the piliferous layer all forming the bark. In the root-thorns, as in the stems, the endodermis and the primary cortex with chloroplasts persist in the old region. Thus the aerial 'root-thorn' differs from the subterranean root in respect of bark formation.

When the root has reached a certain age, most of the cells which contained the greenish yellow granular substance in the young stage, develop tannin. One finds such tannin-bearing cells distributed all over the section from the phloem to the phellogen.

Physiological Study.

(i) *Growth-rate:*

So long as the rains last (till about the beginning of November), the 'root-thorns' of the current year continue to grow and the rate of growth ordinarily is almost the same every day and is about 0·05 cm. a day. It may be noted here that these 'root-thorns' appear mostly on the side of the trunk of the tree along which rain water trickles down in greater quantity and which in consequence is more moist than the other. At the end of the rains the growth stops, probably due to the fall in the humidity of the atmosphere, the tip hardens and the roots are converted into thorns (Pl. I, fig. 3). The length of such thorns ranges from 2·5 to 6 cm. Later, when the stem grows in thickness due to the formation of secondary tissues the older 'root-thorns' appear shorter and shorter as the basal portions are buried in the secondary tissues of the stem. At the same time, the basal region of the old 'root-thorns' also grows in thickness. The pressure exerted by the thickening thorns throws the surrounding secondary tissues of the stem into concentric folds (Pl. I, fig. 2).

(ii) *Factors controlling growth-rate:*

An attempt was made to analyse the factors controlling the growth of these 'root-thorns'.

(a) A 'root-thorn' of the current year was covered with a darkened test-tube but no provision was made for moisture supply. The bottom of the tube had been broken and covered loosely with black paper so as to cut off light without affecting ventilation. The rate of growth was slightly greater than in an ordinary 'root-thorn' and was about 0·075 cm. a day. Thus, its seems that darkness promotes growth, though slightly.

(b) Another young 'root-thorn', growing about two feet above the surface of the soil, was just covered with sand in a box

raised to that height, while others growing at the same level were left exposed. The rate of growth in the covered 'root-thorn' was measured at intervals by uncovering it. In this 'root-thorn', the rate of growth was found to be 0·4 cm. a day. In this case the 'root-thorn' was not only darkened but also received a greater amount of moisture in the sand than in the atmosphere. We thus see that when darkness is combined with increased moisture, the rate of growth is still more increased. This experiment could not be continued longer owing to the limited space in the sand box. 'Root-thorns' at the same height, but uncovered, continued growing at the normal rate, i.e., 0·05 cm. a day.

(c) Yet another 'root-thorn' growing very close to the ground was covered by putting earth over it. The rate of growth in this 'root-thorn' went on increasing in a progression of about 0·2 cm. every two days during the period of observation. Later on, this 'root-thorn' was observed spreading like an ordinary side-root. In this case the 'root-thorn' could secure all the conditions of a subterranean root, i.e., darkness and optimum amount of moisture and hence showed continuous growth like an ordinary root. A 'root-thorn' of the previous year which was by its side but slightly at a higher level, had hardened in the normal course into a thorn, showing that it could not of its own accord grow into the soil.

The following table shows the growth-rate under different conditions:

Day.	Growth-rate in an ordinary root-thorn.	Growth-rate when darkened in the test-tube. (a)	Growth-rate when covered with sand in raised box. (b)	Growth-rate when covered with soil. (c)
3rd.	0·10 cm.	0·12 cm.	0·8 cm.	0·9 cm.
5th.	0·10 cm.	0·13 cm.	0·8 cm.	1·1 cm.
7th.	0·13 cm.	0·13 cm.	0·8 cm.	1·5 cm.
9th.	0·10 cm.	0·14 cm.	0·8 cm.	1·9 cm.
11th.	0·10 cm.	0·15 cm.	0·8 cm.	2·5 cm.
13th.	0·10 cm.	0·15 cm.	0·8 cm.	3·5 cm.

All the 'root-thorns' referred to above are apogeotropic and it is yet to be seen why they are so.

The internal structure of the 'root-thorn' which was made to grow into the soil has not been studied. But the internal structure of a 'root-thorn' after growing for about a fortnight inside the moist sand conforms to the internal structure of an ordinary 'root-thorn' (Pl. II, fig. 4). It is interesting to note further that the branches of the aerial 'root-thorns' are just like the main thorns in every respect.

(d) When the sandbox referred to in (b) was removed after five months, a 'root-thorn' was found growing in the space between the box and the stem. This adventitious root had grown for some distance at right-angles to the stem and then on coming in contact with the exterior of the box had bent down almost vertically into the soil which was propping up the box. This bending had evidently been caused first by contact and then probably gravity came into play. The root had produced four branches and a number of fibrous roots (text-fig. 1).
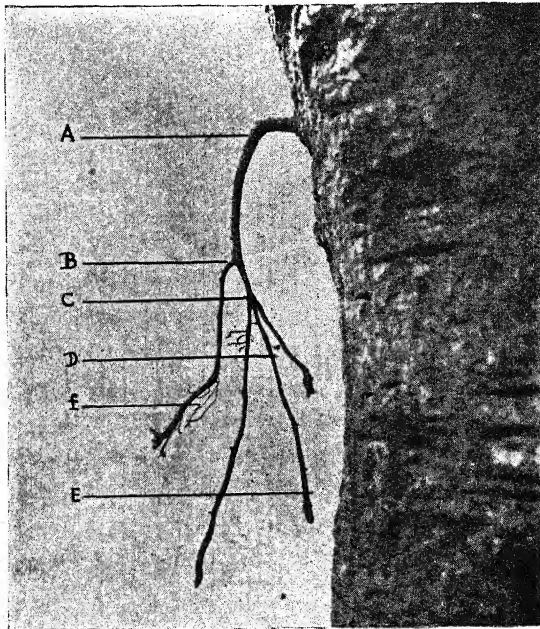
Text-figure 1: Photograph showing a 'root-thorn' (20″ above the ground-level) bending down after coming in contact with the exterior of a sand-box; at A the root bending down due to contact; at B the first branch-root coming out; at C three equally developed branches; f = fibrous roots. × 2/5.

Sections were taken at A, B, C, D, and E. The number of bundles at these levels were about 20 at A, about 16 at B, about 12 at C, 8 at D and 7 at E. It is clear, therefore, that an adventitious root destined to be a thorn, does not change its polystelic character even if it grows down into the soil. But the fibrous roots produced on them possess four xylem bundles in the stele like the ordinary soil roots.

Summary.

1. 'Root-thorns' in *Bridelia pubescens*, Kurz., are recorded probably for the first time· That the thorns are morphologically roots is established by the study of their external and internal structure.

2. As a result of the morphological and physiological investigations the following conclusions are drawn:

(a) The young 'root-thorn' is polyarch like a monocotyledonous root, the outermost layer of cells of the cortex looks like an exodermis; and the epiblems is many-layered and looks like a velamen. It has also a root-cap at its apex.

(b) Secondary growth in the stele is like that of an ordinary dicotyledonous root but the formation of phellogen and bark is like that of a stem. When more secondary growth takes place in the stem the older 'root-thorns' appear shorter and shorter, as their basal region gets buried in the stem tissues.

(c) The 'root-thorns' are apogeotropic and they grow as long as the atmosphere is humid.

(d) If, however, they are darkened, the growth-rate increases slightly and when covered with soil, the rate is greatly increased. Light appears to have no effect on the direction of growth.

(e) When the 'root-thorns' are covered with moist soil while still young, they grow rapidly but in a horizontal direction.

Explanation of Plate I.

Fig. 1. *Bridelia pubescens* tree growing in the back ground of the Ravenshaw College Botanical Laboratory. The two branches on the left have the 'root-thorns' growing on them.

Fig. 2. *Ibid:* A portion of the stem showing concentric folds round an old 'root-thorn'.

Fig. 3. *Ibid:* A portion of the stem with a couple of comparatively young 'root-thorns'.

Fig. 4. *Ibid:* A longitudinal section of the tip of a young 'root-thorn'.

Explanation of Plate II.

Fig. 1. *Bridelia pubescens:* Transverse section of a subterranean old root showing secondary growth. $\times_1$=Primary xylem; $\times_2$=Secondary xylem; Ph_2=Secondary phloem with stereome (S); P=Phellogen; C=Cork; B=Bark.

Fig. 2. *Ibid:* Transverse section of a young subterranean root.

Fig. 3. *Ibid:* Transverse section of an aerial 'root-thorn' showing the commencement of secondary growth. Cb=Cambium.

Fig. 4. *Ibid:* Transverse section of an aerial young 'root-thorn' showing primary xylem bundles.

Fig. 5. *Ibid:* Transverse section of the same showing the many-layered dermal tissue looking like velamen and the delimiting layer (Dl) of the cortex analogous to exodermis.

Fig. 6. *Ibid:* Transverse section of an old aerial 'root-thorn' showing secondary xylem.

Fig. 7. *Ibid:* Transverse section of the same showing the development of phellogen (P).
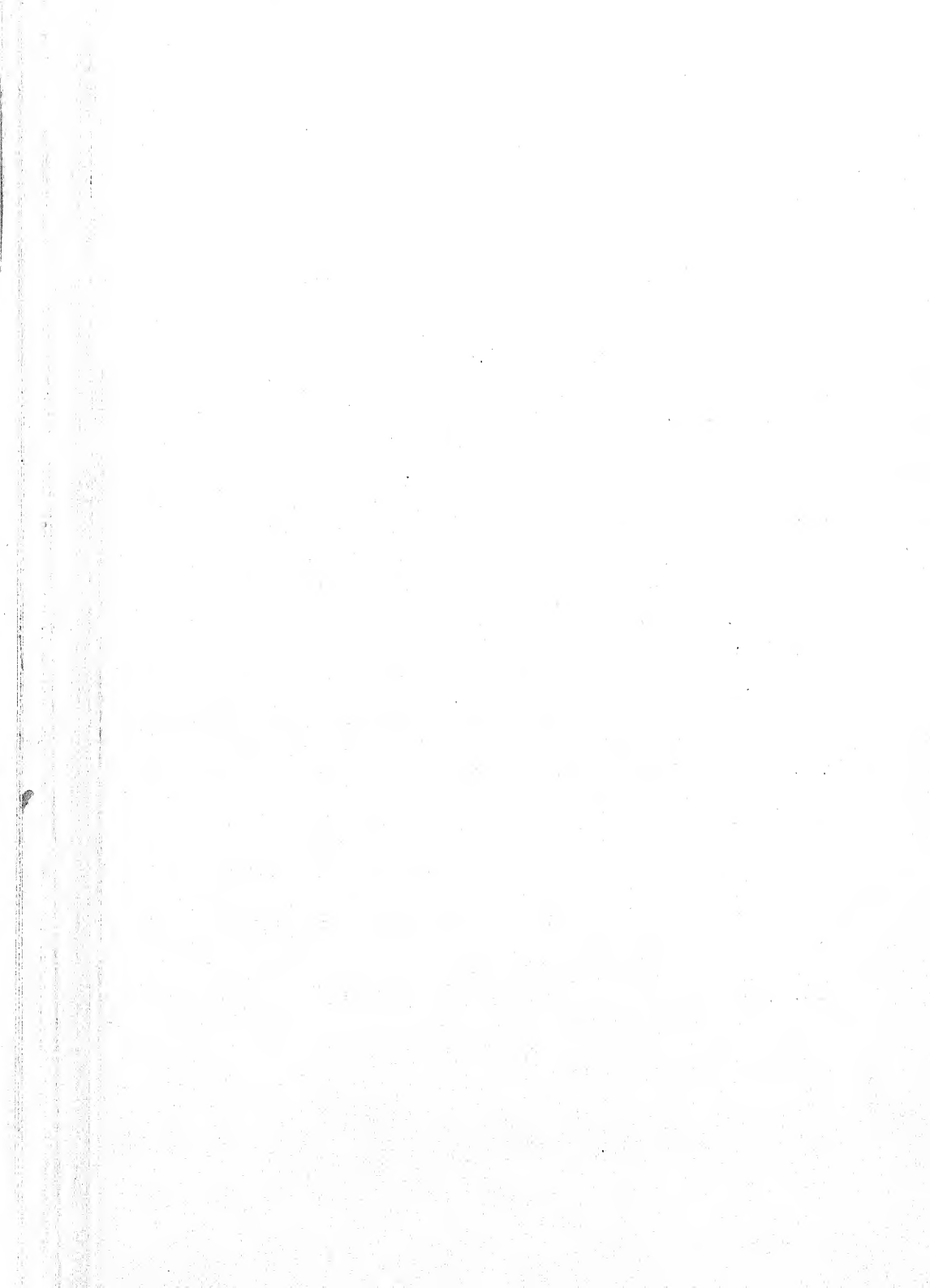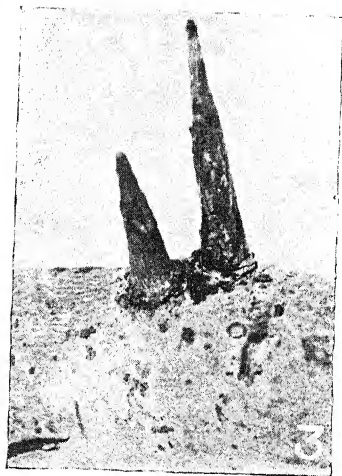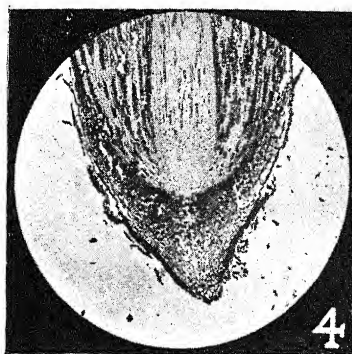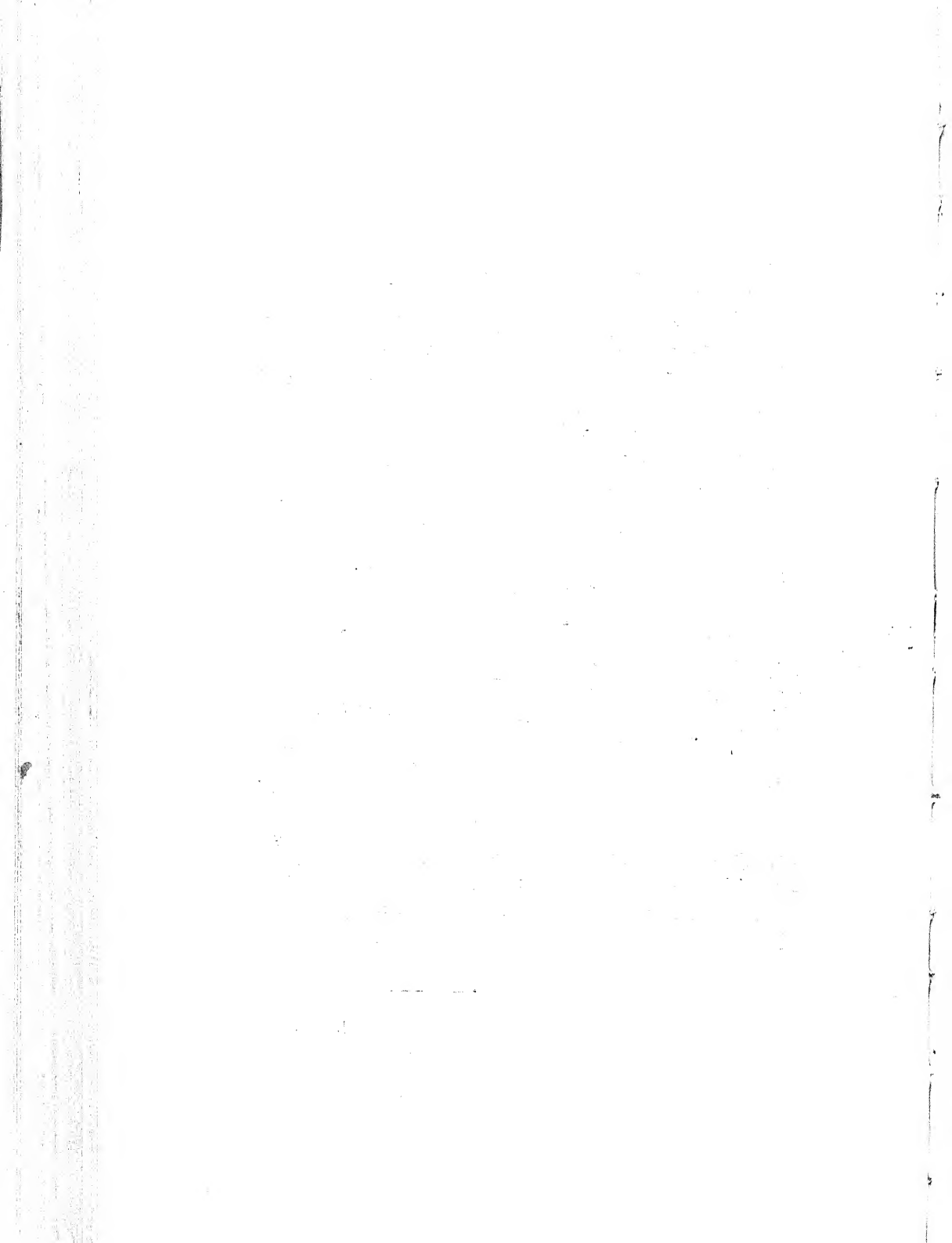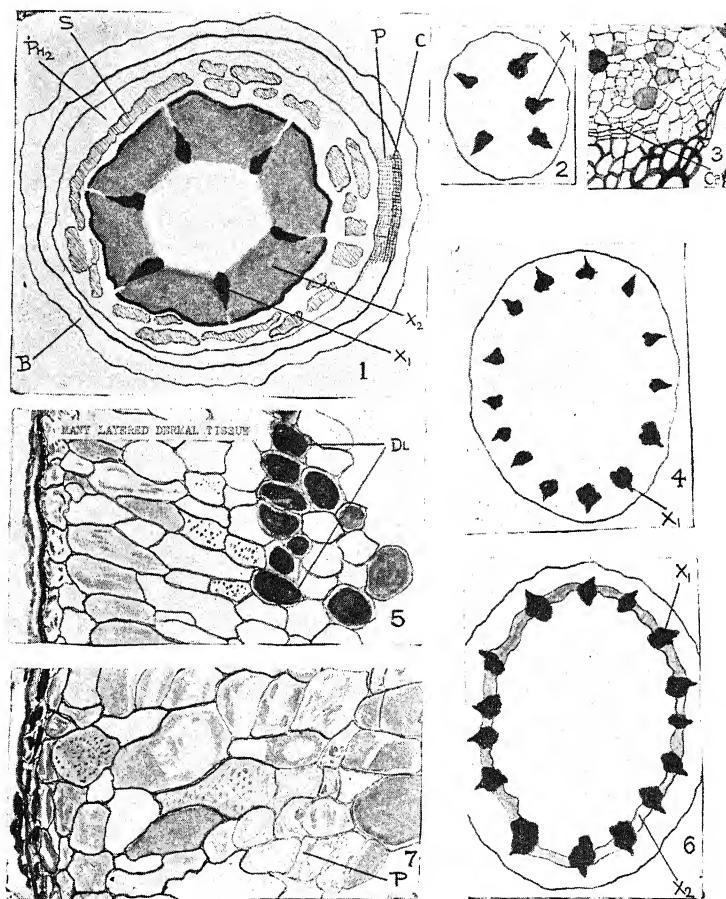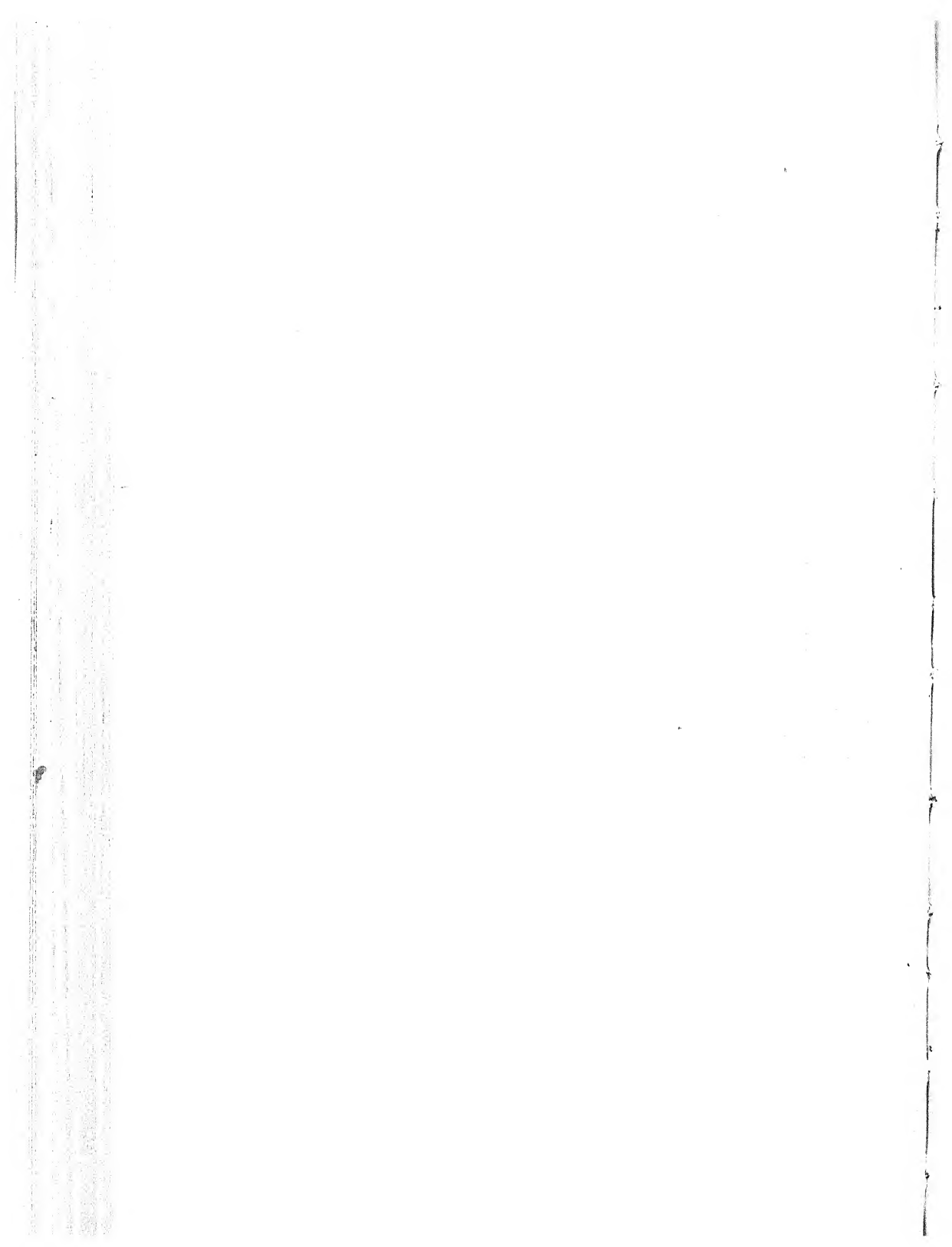
Plate II.

OBSERVATIONS ON THE BIOLOGY AND PHYSIOLOGICAL ANATOMY OF SOME INDIAN HALOPHYTES

BY

D. P. MULLAN,

St. Xavier's College, Bombay.

(Continued from page 179 of Vol. XII, No. 2).

Leucas aspera, Spreng. (Labiatae).

Leucas aspera, Spreng. comes up during the rainy season, covering large tracts of the sandy sea-shore.

The young stems are thrown into ridges and furrows (Fig. 198), while the mature parts are quadrangular. The epidermis covering the ridges or the angles is composed of thick-walled, narrow, vertically-elongated cells (Fig. 199); while that covering the grooves or the flat sides consists of thin-walled, wide, parenchymatous cells (Fig. 200). Clothing hairs are mainly confined to the angles. The trichomes are uniseriate and are seated upon pedestals formed by the dilatation of the epidermal cells (Figs. 201, 202). Glandular hairs, similar in structure to those of the previous plants (16), are present on the stem. Each gland consists of a small stalk-cell and a spheroidal or ovoid head which is divided by vertical walls into four cells (Figs. 200, 203). In the young parts, the glands are more numerous and larger, the head being composed of eight cells. The cuticle, especially in the region of the ribs, is thickly developed and its outer surface is coated with wax. In the young parts, the outer cortex is composed of alternating, vertical strands of collenchyma and chlorenchyma, the former occurring in the projected parts, whereas the latter is more or less sheltered by being placed in the shallow grooves (Fig. 204). Stomata occur in the region of the grooves. The guard cells have thick walls and prominent outer cuticular ridges. In the young parts, the trichomes roof over the regions of the stomata (Fig. 198). In the old parts, the cells of the epidermis and those of the cortex dilate considerably and seem to act mainly as the aqueous tissue. The endodermis shows Caspary's dots. Oil occurs in the cortex of the young stem (Fig. 203 B). The secondary xylem develops more vigorously towards

the four corners of the stem. The pith gets lignified in the old parts. Oxalate of lime occurs in the form of simple or minute needle-shaped crystals or as crystal-sand.

In the leaf, uniseriate trichomes occur on both surfaces, being more numerous on the lower surface (Fig. 205). As a rule, the trichomes on the upper surface are longer and are inserted in an elevated socket formed by the epidermal cells (Fig. 206). Depressed glandular hairs, resembling those on the stem, occur on both surfaces, being more numerous on the lower (Fig. 207). The lamina is 0·27 mm. thick, the upper epidermis being 0·06 mm. The latter seems to act as the main aqueous tissue (15). Stomata occur on both surfaces, being about 150 on the upper and about 200 on the lower (Figs. 208, 209). They are sunk in pits and the guard cells have thick walls and strongly developed outer cuticular ridges (Figs. 210, 211). The leaf is bifacial, consisting of a single layer of palisade tissue and 3–4 layers of closely-packed spongy tissue. Oil and minute rod-shaped crystals occur in the mesophyll.

Boerhaavia diffusa, Linn. (Nyctaginaceae).

The *Nyctaginaceae* is represented on the sandy sea-shore by *Boerhaavia diffusa*, Linn. which also occurs farther inland.

The epidermis of the stem appears opaque, in surface view, due to the deposition of crystalline granules of oxalate of lime. On dissolving away the granules, the cells are seen to hold large nuclei (Fig. 212). The young parts are covered with uniseriate glandular hairs (Fig. 213) whose lower cells hold brownish contents, while the ellipsoidal head is mostly colourless. Stomata occur on the stem. The epidermal cells on either side of the guard cells are thick-walled and arch over on either side of the stoma, hiding the latter in surface view (Fig. 214 A). As is seen in a cross section (Fig. 214 B), the stoma is more or less on the same level as the other epidermal cells, but by the arching over of the surrounding cells a deep, narrow chamber is formed over the stoma, thus producing the same effect as if the latter was depressed. The arching over is also repeated towards the inner face of the stoma. The outer cuticular ridges of the guard cells are prominently formed. Besides these means for reducing transpiration, the respiratory cavity is occluded by the intrusion of a thick-walled hypodermal cell (*h*). The hypodermal and the cortical cells are clear (Fig. 215). Vertically-elongated crystal cells, holding raphides, occur in the cortex (Fig. 216). The endodermis holds chloroplasts which line the inner walls. The anomaly, in *B. diffusa*, has been investigated by Maheshwari (14).

The phellogen layer arises subepidermally or in a deeper layer of the cortex (Fig. 217). The basal part of the stem acts as a storage organ: the cells of the cortex, the conjunctive tissue and the pith being fully loaded with starch.

In the petiole, glandular hairs occur on the abaxial surface. A few stomata are confined to the sides of the petiole where the respiratory cavity is occluded as in the stem. Heimerl (10) has demonstrated the deposition of crystalline granules of oxalate of lime in the walls of the epidermal cells of the leaves of *Boerhaavia*. In *B. diffusa*, the crystalline deposition is more pronounced on the undersurface of the leaf. Uniseriate glandular hairs arise on both surfaces, being more numerous on the lower. The majority of hairs are short (Fig. 218), while the long ones are mainly confined to the lower surface (Fig. 219). The hairs resemble, in structure and contents, those of the stem. Stomata are equally abundant on both surfaces (Figs. 220, 221), being about 150 per sq. mm. in the halophytic form; while in inland forms, they are about 100—the apparent increase in the halophytic form being due to the reduction of the leaf surface. The characteristic arching epidermal cells of the stem do not accompany the stomata of the leaf (Fig. 222), but instead, the cuticle around each stoma arches outwards and forms a shallow pit (Figs. 223, 224). The latter is filled up with crystalline granules, thus clogging the outer stomatal opening. The outer as well as the inner cuticular ridges of the guard cells are prominently developed and closely approximated. The respiratory cavities are reduced on both surfaces. The cuticle is thickly developed, being more pronounced on the lower surface. The epidermal cells on both surfaces are deep and seem to act as the aqueous tissue(15). The lamina is 0·37 mm. thick, each epidermis being 0·06 mm. The leaf structure is bifacial. After the monsoon, the palisade cells of the old leaves get somewhat dilated and seem to act as subsidiary water-reservoirs. Small aggregate crystals arise at the upper ends of the palisade cells. The latter are interrupted, at intervals, by idioblasts (resembling in outline the palisade cells) holding raphides. Tannin-bearing cells also occur in the palisade tissue. The lowermost spongy cells are dilated and more or less clear (Fig. 224). This layer seems to correspond to the aqueous tissue noted by Sabnis (19) in a xerophytic form of the plant. The veins are all invested by a sheath of thick-walled, closely-fitting, more or less funnel-shaped cells holding large, immobile

chloroplasts (with included starch) at their narrow, basal ends
(Figs. 225, 226). The spongy tissue is traversed by elongated
idioblasts (holding raphides) which run parallel to the leaf sur-
face.

The root also shows anomalous growth. Crystal cells, hold-
ing raphides, occur in the cortex. The primary root acts mainly
as a storage organ, being fully loaded with starch, and attains a
diameter of 10-15 mm.

In plants growing inland, the glandular hairs on the young
stem are few in number and are mostly devoid of contents. In
the stem, the cells accompanying the stomata form a comparative-
ly wider chamber (Fig. 227). Furthermore, the respiratory
cavity is not occluded by the intrusion of a hypodermal cell and
the outer cuticular ridges of the guard cells are not closely ap-
proximated. The chlorenchyma is fully developed in the stem
and leaf. In the leaf, the lower epidermis shows the typically
mesophytic characteristic in the irregularly wavy lateral walls
(Fig. 228). In the stomata, the outer openings are not clogged
by crystalline granules and the cuticular ridges are not promi-
nently developed (Fig. 229).

Celosia argentea, Linn. (Amaranthaceae).

Celosia argentea, Linn. is found growing in different soils
and under varying conditions. The plant comes up during the
rains and is often found on the sea-shore. Like the previous
plants, the shoot of the maritime form of the plant is quite stunted
in growth (Fig. 230), while that of the inland form is very luxuri-
antly developed (Fig. 231).

The stem of the halophytic form is grooved. The epidermal
cells, as well as the outer cortical cells, vary according as they
form part of the groove or the ridge. The epidermis of the ridges
is composed of thick-walled cells, which are elongated in the direc-
tion of the long axis of the stem (Fig. 232); while the cells of
the groove are thin-walled and appear somewhat polygonal in
surface view (Fig. 233). The cuticle is thickly developed on the
ridges. Stomata are confined to the grooves and the guard cells
have thick, outer cuticular ridges (Fig. 234). The ridges of the
stem are stiffened by collenchyma, while, in the region of the
groove, the epidermis is followed by a single layer of chlorenchy-
ma, consisting of thin-walled, palisade-like cells (Figs. 234, 235).
Thus the collenchyma and the chlorenchyma form alternating
strands in the young stem. Aggregate crystals and coarse

granules of oxalate of lime occur in the cortex. The nature of
the anomalous structure of the stem has been demonstrated by
de Bary (1) and by Solereder (23).

In the petiole, the chlorenchyma is confined to the two sides
and consists of 2–3 layers of loosely-arranged palisade-like cells
(Fig. 236). Stomata are more numerous in the region of the
chlorenchyma. Large crystal cells, holding crystal-sand, occur
towards the under side of the main vein and at the sides of the
petiole (Fig. 237). Stomata occur on both surfaces being 75
on the upper and 200–225 on the lower (Figs. 238, 239). The
apparent large number of stomata is again due to the great reduc-
tion of the leaf surface in the maritime form. The leaf structure
inclines more towards the isolateral type, the mesophyll tissue
being composed of more or less oblong cells (Figs. 240, 241). The
minor veins are surrounded by a sheath of large, clear, parenchy-
matous cells (Fig. 242). At times, the cells (k) of the lower
face of the veins hold crystal-sand. The old leaves become thick
and succulent and assume a pale yellow colour. The lamina
of the functional leaf is 0·64 mm. thick, while in the old leaves
it is nearly 1·5 mm., being thicker towards the margin. The
thickness of the old leaves is seen to be due to the dilatation of
the mesophyll cells (Figs. 243, 244). The chlorophyll content
of the old leaves is poor. A comparison of figures 240 and 241
with Figs. 243 and 244 (all magnified equally) brings out clearly
the extent of the dilatation of the mesophyll cells in the case of
the old leaves as compared to those of the functional leaves.
Thus the old leaves act mainly as water reservoirs and resemble
the old leaves of some of the previous plants (15).

The internal anatomy of the plants growing inland shows
several mesophytic characters. The stem is more prominently
grooved, the collenchyma being more fully developed. The assi-
milatory tissue is very prominently developed in the stem, petiole
(Fig. 245) and leaf. The lamina is thin, being one-half that of
the halophytic form. In the mesophytic form, the lower epider-
mal cells have more wavy lateral walls (Fig. 246) and the leaf
structure is typically bifacial (Fig. 247).

Sesuvium portulacastrum, Linn. (Ficoideae).

Sesuvium portulacastrum, Linn. is one of the few succulent
xerophytes growing on the sea-shore, either on the verge of the
high-water mark, or on the dry sands. The stout, fleshy stems
creep along the sands, rooting at the nodes (Fig. 248). This

4

habit facilitates vegetative multiplication, for, when parts of the stem get separated, *e.g.*, by the action of the tides, they are able to take root and establish themselves when washed ashore.

The anatomical structure is typical of the succulents. The cuticle of the stem is well developed and impregnated with mucilage. The outer cortex is composed of vertically elongated cells. At times, the intercellular spaces of the cortex are seen, in preserved material, to hold a yellowish substance which proves to be mainly mucilage (Fig. 249). Anthocyanin occurs in the epidermis and in the outer cortex of the exposed parts. A few chloroplasts and starch grains occur in the cortical cells. The inner cortex becomes lacunar by the cells getting tangentially stretched and developing schizogenous cavities (Fig. 250). Regnault (18) and Solereder (24) have noted an anomaly in the rising of successive arcs of meristem outside the first normal ring of vascular bundles. Starch occurs in the medullary rays and in the pith. Crystal cells, with aggregate crystals, are present in the cortex and the pith. Cork originates in the region of the lenticels which arise on the basal part of the stems.

The leaves vary in shape on different soils, being mostly linear or spathulate-oblong. On dry soil and during the hot season, they are reduced and tend to become sub-cylindric. Solereder (24) and others have noted the occurrence of large, bladder-like, water-storing cells in the epidermis of *S. portulacastrum*. In the individuals growing on the sea-shore, no such cells are found. Oil is present in the epidermis (Fig. 252) and anthocyanin occurs in the cells at the apex and the margins. Stomata are equally abundant on both surfaces, being 75 per sq. mm. (Fig. 251). The guard cells have thick walls and the outer cuticular ridges are thickly formed (Fig. 252). The cuticle is impregnated with mucilage. The leaf structure is isolateral and consists of a centrally-placed aqueous tissue which is surrounded by 3–4 layers of palisade cells (Figs. 253, 254). Towards the lower surface, the palisade tissue is less prominently developed. In a lamina 2.4 mm. thick, the aqueous tissue occupies 1·5 mm. Except for the presence of a few chloroplasts and starch grains, the cells of the aqueous tissue are clear. Crystal cells, with aggregate crystals, occur on the inner face of the palisade tissue. These crystal cells are peculiar in that they hold chloroplasts which are arranged around the inner walls (Fig. 254, *k*). Wound cork develops on the lamina whenever it gets injured.

The cortex of the root shows schizogenous lacunae (Fig. 255). The anomaly in the root is brought about by arcs of meristem arising in the pericycle and producing secondary bundles and conjunctive tissue. The secondary bundles arise at first on either side of the primary bundles which form an axile strand (Fig. 256 A). As growth proceeds, fresh bundles are added all round the primary (x_1) strand (Fig. 256 B). The secondary bundles (x_2) ultimately fuse and form a ring (Fig. 256 C). The same process is then repeated on either side of the first ring of secondary tissue by the arising of arcs of meristem. Later, fresh bundles are added which also fuse and form a second ring (x_3) around the first one (Fig. 256 D), and so on. Starch occurs in the cortex and in the conjunctive tissue. Cork (k) arises superficially.

Suaeda nudiflora, Moq. (Chenopodiaceae).

Suaeda nudiflora, Moq. is another succulent xerophyte which is often found growing near the sea. During the dry season, the leaves assume a semi-terete form, thereby reducing the evaporating surface.

The internal structure shows many of the peculiarities of the succulents. The young stems are soft and fleshy, while the older parts are woody. The cuticle is not prominently developed. A few stomata are present on the young parts. The stomata are abnormally placed, with their openings arranged transversely to the long axis of the stem (Fig. 257). The epidermal cells surrounding each stoma are raised above the general surface of the stem (Fig. 258). The epidermis is succeeded by a layer of more or less collenchymatous cells with the thickenings at the angles pierced. The rest of the cortex consists of thin-walled, vertically-elongated cells. In preserved material, the intercellular spaces of the cortex are seen to hold a yellowish substance which proves to be mainly mucilage (Fig. 259). Oil occurs in the epidermis and the cortex. Crystal-sand and aggregate crystals of oxalate of lime are present in the cortex. Anthocyanin is mainly confined to the endodermis, though it also occurs, here and there, in the epidermis and hypodermis of the exposed parts. A peculiarity of the young stem is the position of the chlorophyll tissue. Very few chloroplasts occur in the cortex; while they are present in the endodermis and are even to be found within the stele, occurring in the pericycle, the conjunctive tissue and towards the inner face of the primary bundle. Secondary thickening leads

to an anomalous growth. The anomaly is produced by successive rings of cambium which arise centrifugally in the pericycle, near the inner margin of the bast fibres. The meristem produces secondary bundles and conjunctive tissue composed of thick-walled, lignified, prosenchymatous cells. In the bundles, secondary xylem is produced internally and soft bast externally. Thus in a cross section, the vascular bundles are seen to be embedded (in a more or less concentric manner) in a thick ring of conjunctive tissue. In the young parts, the pith consists of thin-walled cells, holding oil and chloroplasts; while in the old parts, it gets pitted and lignified. The phellogen layer arises in the pericycle (Fig. 260).

The thickness and shape of the leaves vary according to the soil. The epidermal cells are deep, evidently acting as water reservoirs. In a leaf 1·7 mm. thick, the epidermis, on either surface, is 0·15 mm. deep. The cuticle is poorly developed and the outer surface is coated with granules of wax. Oil occurs in the epidermis. Stomata are small, few and equally distributed on both surfaces, being about 50 per sq. mm. (Figs. 261, 262). Solereder (24) has noted the occurrence of subsidiary cells, placed parallel to the pore, in the leaves of *S. fruticosa*. In *S. nudiflora*, similar cells occur, but mostly on one side of the guard cells (Fig. 262) and that too not in all cases. The stomata are abnormal in that the pores are placed transversely to the direction of the median vein. They are slightly depressed and have small, outer, cuticular ridges (Fig. 263). The leaf structure of *S. nudiflora* is isolateral (Fig. 264) and resembles that of *S. Forskallii*, Solms., *S. pruinosa*, Lge· and *S. vermiculata*, Forsk. described by Solms-Laubach (25). The palisade tissue consists of a single layer of short cells which is followed by collecting cells (Figs. 265, 266). The hollow cylinder formed by these assimilatory cells is filled up by a central, aqueous tissue (*a*) consisting of large, thin-walled cells which are mostly elongated at right angles to the leaf surface. In a leaf 1·7 mm. thick, the aqueous tissue is seen to occupy 1·3 mm. The veins are embedded in the aqueous tissue. The old leaves have a tendency to turn yellow and become thick (Fig. 267). Such leaves seem to act mainly as water reservoirs like the old leaves of some of the previous plants (15). They get more than twice as thick as the functional leaves, the thickness being due to the dilatation of the aqueous tissue.

The anomalous structure of the stem is repeated by the root. A peculiarity of the anomalous growth, as described by Fron (6)

in the various species of *Suaeda,* is that the vascular bundles exhibit a spiral growth (Fig. 268).

Arthrocnemum iudicum, Moq. (Chenopodiaceae).

Arthrocnemum indicum, Moq. is an aphyllous succulent which is found growing on wet, saline soil. The plant is an annual and dies down at the end of the dry season, after producing seeds which germinate on the advent of the rains. The stems are fleshy and become woody towards the base.

The cuticle is feebly developed and the outer surface is striated. The stomata (Fig. 269) are few and are abnormally arranged, as in the last plant. They are even with the surface and the guard cells have thick walls and prominent outer cuticular ridges (Fig. 270). The epidermis is followed by 2–3 layers of palisade-like cells some of which hold oil. This hollow cylinder of chlorophyll tissue is filled with a prominently-developed aqueous tissue composed of large, parenchymatous cells (Figs. 271, 272). Sphaero-crystals and crystal-sand occur in the aqueous tissue. The latter is supported on its outer face, *i.e.* just beneath the palisade cells, by a network of feebly-lignified vascular tissue, the xylem part of which points inwards and consists of spiral and pitted elements. In *A. indicum,* Duval-Jouve (4) and Dangeard (3) have noted the absence of sac-like tracheides which occur in some species of *Arthrocnemum.* The stele is contracted. Sabnis (3) has noted an anomalous structure in *A. indicum.* Irregular crystals of oxalate of lime occur in the pith. The phellogen layer arises in the pericycle, immediately internal to the endodermis. Hence, at the basal part of the stem, the assimilatory and the aqueous tissues are thrown off.

In the root, the primary cortex is lacunar (Fig. 273). The cortical cells are somewhat vertically elongated and the lacunae are schizogenous in origin. From its lacunar cortex it is seen that *A. indicum* is adapted, like the plants of Part I, to living in the badly-aerated mud near the sea. The anomalous structure of the stem is repeated by the root. The greater portion of the root is composed of conjunctive tissue in which the secondary vascular bundles are embedded more or less concentrically. Cork develops superficially and is composed of lignified, phelloid cells — again, a characteristic feature of the plants of Part I.

Seedlings of *A. indicum* were removed from their natural habitat and grown under mesophytic conditions for nearly seven months. In the halophytic form, the branches remain close to the main

stem, thus pointing their apices to the sky; while, under mesophytic conditions, they spread out more or less horizontally. In the stem of the mesophytic form, the chlorophyll content is more abundant, giving the branches a bright green colour. The epidermal cells are less deep and the palisade tissue is more loosely arranged (Fig. 274). The network of vascular tissue is composed of wider elements. The endodermis, which is clear in the halophytic form, shows the presence of starch grains. The aqueous tissue gets reduced (Fig. 275), with the consequence that, under mesophytic conditions, the branches are thinner than those of the halophytic form.

Discussion

Of the two groups of halophytes, the plants of the salt swamp are all perennial evergreen shrubs or trees with thick, leathery leaves. The seeds or seedlings of the mangroves are mainly dispersed during the monsoon. In their internal structure the plants show a remarkable general similarity. In the stem, the inner cortex, as a rule, develops schizogenous lacunae. The otherwise soft cortex is strengthened by sclerenchymatous elements in the form of long, H-shaped spicules or multiradiate sclereides or stone-cells. In most stems, the secondary cortex gets differentiated as an aerating tissue. The pith, at maturity, gets pitted and lignified, thus adding to the mechanical strength of the stem. Since the pith also acts as a storage tissue, the possession of pits is advantageous, for, as Haberlandt (9) has pointed out, it facilitates an active interchange of materials between the cells and with those of the contiguous conducting parenchyma. In the leaf, there is a strongly developed cuticle whose protective function is further augmented by a coating of wax. Stomata are prominently depressed and the outer cuticular ridges of the guard cells are thickly developed and form a distinct hyperstomatic chamber. In some cases, the efficiency of the outer vestibule is further increased by the splitting of the cuticular ridges which thus form a double chamber over the stoma proper. In all cases, save one, the stomata are confined to the undersurface of the leaf and are, on an average, about 129 per sq. mm. The leaves of the halophytic forms in general are thicker and more succulent than those of the mesophytic forms of the same plants. Lesage (13) has proved that the presence of salts in the soil is the main cause of succulency in the maritime plants. The leaves of the man-

groves are much thicker than those of the psammophilous halo-
phytes and the thickness is seen to be due mainly to a specially
developed aqueous tissue. With the exception of one individual,
the water-tissue occurs beneath the upper epidermis of the leaf
and, in a few cases, a similar, but less prominent, tissue also occurs
beneath the lower epidermis. On an average, the leaves are 0·55
mm. thick, while the subepidermal aqueous tissue occupies 0·19
mm. of the total thickness. Schimper (22) regards the aqueous
tissue of the halophytes as: specially adapted to guard against
the injurious concentration of salt in the assimilating cells; while
Warming (27) inclines to the opinion that a peripheral aqueous
tissue, in addition to acting as a water reservoir, serves to check
the penetration of heat rays and thereby retards transpiration.
According to Faber (5) the water-tissue of the mangroves is a
"cataplastic hypertrophy", caused by the high osmotic pressure
of the cells. Under mesophytic conditions, the aqueous tissue
shows a tendency towards reduction (15). A marked peculiarity
of the leaves of several plants is the occurrence of cork-warts
which may be a means of reducing transpiration. The glandular
hairs of the mangroves resemble the salt-glands of some desert
plants.   Incrustations of hygroscopic salts were observed on the
leaves of several mangroves (16). Keller (12) regards the ex-
cretions of easily soluble salts as an external osmotic mechanism
which sucks water from the interior of the plant; while Faber (5)
is of opinion that it prevents too great a concentration of salt in
the leaf tissue. According to Schimper (22) the xerophilous
structure of the halophytes is directly due to the salt content of
the substratum which makes the latter physiologically dry. The
investigations of Faber (5) have shown that the theory of physi-
ological drought rests on slender evidence. The latter author is
of opinion that the mangroves are facultative halophytes showing
comparatively weak xerophytic characters.

     The most striking modification in the root of the mangroves
is the development of an aerating system which, in some cases, is
further elaborated by the production of pneumatophores. This
aerating tissue does not originate in the phellogen, as is the case
in several marsh plants (20), but is formed by the primary cortex.
The latter is composed of a loose parenchyma whose cells, in many
cases, are radially stretched and hang together by short, lateral
arms. The prominent cortical lacunae evidently serve for the
transport of oxygen from the air, either through the pneumato-
phores or through the lenticels which are thickly developed on the

basal part of the stem. Even when the plants are grown out of the salt swamp, the lacunar cortex persists—thus giving support to Goebel's (8) view that it is not a variable, reaction structure, but is a fixed congenital one. A characteristic of the lacunar cortex is the occurrence of strengthening ridges which form a pretty coherent, feebly-lignified network. The ridges are elastic and seem to serve as a device against side pressure, as suggested by Warming (28). In cases where the ridges are absent, their places are taken by peculiar thickening plates with annular slits. In several cases, the cortical tissue is further supported by multiradiate sclereides, which seem to act like the internal hairs of the aquatic and marsh plants. Karsten (11), Goebel (7) and Schenck (21) have shown that the function of the pneumatophores is to supply the submerged parts with oxygen for their respiration. In cases where the breathing roots are absent, the abundant development of lenticels and of cortical lacunae in the epigeous parts of the stilt-roots or on the basal part of the stem, seems to suggest, as is shown by Goebel (8) in the case of marsh plants, that these parts help in the exchange of gases for the submerged roots. The structure of the pneumatophores, and of the aerial parts of the stilt-roots, inclines more towards that of the stem. Thus the stele, which is contracted in the terrestrial root, expands in the epigeous parts; and sclereides, when present, occur more abundantly in the part which is in the air than that which is buried in the mud. In both the stem and the root of the mangroves, the cork is not of the usual suberised type but consists of lignified, phelloid cells.

Of the psammophilous halophytes, the annuals show less xerophytic characteristics than do the perennials. Since the former live only during the monsoon, they are not faced with the same difficulty as are the perennials. The intense insolation that prevails on the sea-shore is much tempered during the cloudy monsoon days. Thus the more equable climate, coupled with a supply of fresh water, seems to determine the less xerophilous structure of the annuals. The perennials, on the other hand, show marked xerophytic characters. The chief modifications of the anatomical structure are again concerned with the storage of water and the reduction of transpiration. Since the leaves, in most cases, are reduced in size and number, the stem region takes up the assimilatory activities. In such cases, the chlorophyll tissue is protected either by being placed in grooves or by being covered by a felting of trichomes. In glabrous stems, the chloroplasts are screened from the intensity of light by being placed in cells which

are elongated at right angles to the outer surface of the stem. That this palisade-like form of the assimilatory cells is determined by the intensity of light, is seen from the fact that they occur only on the exposed side of the stem. Furthermore, the palisade-like cells are seen to develop in the halophytic forms, while, in the mesophytic forms of the same plants, the chlorophyll tissue is composed of roundish-polygonal cells. As in the mangroves, the pith, at maturity, gets pitted and lignified. The leaves are either small and crowded together or assume the profile position. The cuticle is well developed and in most cases covered with wax. In the majority of the sand plants, stomata occur on both surfaces and are, on an average, 83 per sq. mm. on the upper and 95 on the lower surface. The stomata are protected by being placed in grooves or pits; and when they are even with the surface, they are covered by trichomes. In a few cases, the cavity of the pit is filled with wax or with crystalline granules which evidently serves to reduce transpiration. The outer cuticular ridges are not so well developed as in the mangroves. Unlike the latter, in the psammophilous halophytes no special aqueous tissue is developed, but the storage of water is taken up by the upper epidermis which consists of deep, clear cells (15). On an average, the leaf is 0·29 mm. thick, the upper epidermis occupying 0·046 mm. of the total thickness. The leaves of several perennials are seen to store up water. This is especially marked in the cases of old leaves which become very thick and succulent. Such leaves have poor chlorophyll content and seem to serve essentially as water reservoirs for the younger leaves (15).

Many of the psammophilous halophytes are also found growing inland. The configuration and anatomical structure of such plants differ in several respects from the individuals growing near the sea. The stunted shoot of the halophytic form develops luxuriantly under mesophytic conditions. Thus the ready access or difficulty in obtaining water and a sheltered or exposed situation are seen to have a marked influence on the growth and development of the two forms. In the inland form, the chlorophyll tissue is abundantly developed. The pith of the stem region remains thin-walled, unpitted and unlignified. A marked characteristic of the inland forms is the modification that takes place in the lower epidermis of the leaf. Under halophytic conditions, the latter is seen, in surface view, to be composed of small cells with straight lateral walls; while, under mesophytic conditions, it consists of large cells with irregularly wavy walls, thus giving a

greater diffusion surface to the substances passing from cell to cell. The leaves in the inland form are thinner and the upper epidermis is less deep (15).

The three succulent halophytes agree in having certain common characteristics of their own. The stomata are not protected in any way and the cuticle is poorly developed. On an average, stomata are 50–75 per sq. mm. The leaf structure is isolateral with a centrally placed aqueous tissue and a comparatively poorly-developed, peripheral chlorophyll tissue.

Of the various cell contents three seem to be pretty constant in a large majority of the coast vegetation. The first one is anthocyanin which is specially developed in those aerial parts which are not covered with hairs. It is related to the position of the part to light and seems to serve as a protective screen against intense illumination. In most plants, it occurs in the epidermal or subepidermal cells, being interposed between the chlorophyll tissue and the light. But when the chlorophyll tissue is deep-seated, *e.g.*, in the stem of *Suaeda nudiflora*, anthocyanin also occurs in an inner layer (endodermis) which abuts upon the chlorophyll tissue. The commonest cell contents are, however, tannin and oil. That tannin is produced for some definite purpose and is not a mere by-product which occurs under all circumstances is seen from the fact that, when the halophytes are cultivated under mesophytic conditions, very little tannin is developed. Warming (26) suggests that tannin, in connection with the hygroscopic capacity of acids, would afford a protection against desiccation; while Faber (5) thinks it probable that tannin may be playing an important part in the generation and regulation of turgor-pressure. In the halophytes, it occurs most abundantly in the aerial parts and is present even in the seedling stage. Like tannin, oil also occurs at an early stage, being present in the seedling. It occurs in the aerial parts and is one of the most conspicuous features of the leaves of the halophytes. Pfeffer (17) suggests that it is possible that the presence of oil increases the power of resistance to desiccation.

Literature Cited.

1. BARY, A. DE: Comparative Anatomy of the vegetative organs of the Phanerogams and Ferns. Engl. Ed., Oxford, 1884, pp. 590–91.

2. BLATTER, E., McCANN, C. & SABNIS, T. S.: The Flora of the Indus Delta. Ind. Bot. Soc. Publication, Madras, 1929, p. 138.

3. DANGEARD: Struct. d. Salicornieae at d. Salsolaceae. Bull. Soc. Bot. d. France, xxxv, 1888, pp. 157; 197 *sqq.*

4. DUVAL-JOUVE: Salicorniées. Bull. Soc. Bot. d. France, 1868, pp. 132–140.

5. FABER, E. C. VON: Zur Physiologie d. Mangroven. Ber. d. Deutsch. Bot. Gesellsch., xli, 1923, pp. 227–234.

6. FRON: Racine d. Suaeda et d. Salsola. Comptes Rendus,, Paris, cxxv, 1897, pp. 366–68; and Struct. Spiralée d. Racines d. cert. Chenopodiae. Comptes Rendus, Paris, cxxvii, 1898, pp. 563–65.

7. GOEBEL, K.: Uber d. Luftwurzeln von Sonneratia. Ber d. Deutsch. Bot. Gesellsch., iv, 1886, pp. 249–55.

8.   „   „   : Pflanzenbiologische Schilderungen. i–ii, Marburg, 1891–93.

9. HABERLANDT, G.: Physiological Plant Anatomy. Engl. Ed., 1928, pp. 44; 329.

10. HEIMERL: Einlag. v. Calcium-Oxalat etc. Sitzungsber. d. Wiener Akad., xciii, 1, 1886.

11. KARSTEN, G.: Uber d. Mangrove-Vegetation im Malayischen Archipel. Biblioth. Bot., xxii, 1891, pp. 46 *sqq.*

12. KELLER, B. VON: Halophyten- und Xerophyten-Studien. Journ. of Ecology, xiii, Cambridge, 1925, pp. 224–61.

13. LESAGE, P.: Researches experimentales s. 1. modifications d. feuilles c. 1. plantes maritimes. Rev. Gén. d. Bot., ii, 1890. pp. 55, 106, 163; see also: Comptes Rendus, cxii, 1891, p. 672.

14. MAHESHWARI, P.: Contributions to the morphology of Boerhaavia diffusa, Journ. Ind. Bot. Soc., ix, 1, 1930, pp. 42–61.

15. MULLAN, D. P.: Observations on the water-storing devices in the leaves of some Indian halophytes. Journ. Ind. Bot. Soc., x, 2, 1931, pp. 126–33.

16.   „   „   : On the Occurrence of glandular hairs (salt glands) on the leaves of some Indian halophytes. Journ. Ind. Bot. Soc., x, 3, 1931, pp. 184–89.

17. PFEFFER, W.: The Physiology of Plants. Engl. Ed., Oxford, i, 1900, p. 595.

18. REGNAULT: Annales d. Sciences Naturelles—Botanique, ser. 4, xiv, 1860, pp. 95–105.

19. SABNIS, T. S.: The physiological anatomy of the plants of the Indian desert. Journ. Ind. Bot. Soc., ii, 1921, pp. 101–2.

20. SCHENCK, H.: Uber d. Aerenchym etc. Pringsch. Jahrb., xx, 1889.

21.   „   „   : Uber d. Luftwurzeln v. Avicennia tomentosa u. Laguncularia racemosa. Flora, lxxii, 1889, pp. 83–8.

22. SCHIMPER, A. F. W.: Plant Geography upon a physiological basis. Engl. Ed., Oxford, 1903, p. 89.

23. SOLEREDER, H.: Holzstruktur. 1885, pp. 141–3, 211–13.

24.   „   „   : Systematic anatomy of the Dicotyledons. Engl. Ed., Oxford, 1908.

25. SOLMS-LAUBACH, H.: Spirolobe Chenopodeen. Bot. Zeitg., i, 1901, pp. 168–70.

26. WARMING, E.: Botanisches Centrablatt. xvi, 1883, p. 350.

27.   „   „   : Oecology of Plants. Engl. Ed., Oxford, 1909, p. 121.

28.   „   „   : Rhizophora Mangle. L. Tropische Fragmente, ii, Engler's Jahrb., iv, 1883, pp. 517–48.

Explanation of Plates.

The initial magnification is indicated after each figure. All the figures have been reduced to about one-third in reproduction.

Leucas aspera, Spreng.

Fig. 198.—T. S. young stem: *c*, collenchyma; *ch*, chlorenchyma. (× 41).

Fig. 199.—Epidermis, stem: *g*, gland; *t*, trichome. (× 500).

Fig. 200.—Epidermis, stem: region of the groove. (× 500).

Fig. 201.—T. S. stem, showing a trichome. (× 240).

Fig. 202.—   do.   do.   (× 500).

Fig. 203.—T. S. stem, showing a gland: *A*, on the ridge; *B*, on the groove. (× 500).

Fig. 204.—T. S. stem: *c*, collenchyma; *ch*, chlorenchyma, (× 500).

Fig. 205.—T. S. leaf, showing a trichome on the lower surface. (× 240).

Fig. 206.—   do.   do.   on the upper surface. (× 240).

Fig. 207.—A gland on the upper surface of the leaf: *A*, in surface view; *B*, in cross section. (× 500).

Fig. 208.—Leaf: upper epidermis; *g*, gland; *t*, trichome. (× 240).

Fig. 209.—Leaf: lower epidermis. (× 240).

Fig. 210.—T. S. leaf: *e*, upper epidermis; *p*, palisade cells with oil. (× 500).

Fig. 211.—T. S. leaf: *l*, lower epidermis; *s*, spongy tissue. (×240).

Boerhaavia diffusa, Linn.

Fig. 212.—Stem: epidermis. (× 240).

Fig. 213.—Stem glandular hair. (× 500).

Fig. 214.—Stem, showing the structure of the stoma: *A*, in surface view; *B*, in cross section. (× 500).

Fig. 215.—T. S. stem: *e*, epidermis; *h*, hypodermis; *n*, endodermis. (× 500).

Fig. 216.—L. S. stem, showing the cortex (× 240).

Fig. 217.—T. S. stem, showing the phellogen layer, *p*, (× 180).

Fig. 218.—Leaf: a glandular hair on the upper surface. (× 500).

Fig. 219.—   do.   do.   on the lower surface: *A*, in surface view; *B*, in cross section. (× 500).

Fig. 220.—Leaf: upper epidermis. (× 240).

Fig. 221.—Leaf: lower epidermis. (× 240).

Fig. 222.—Leaf: lower epidermis. (× 500).

Fig. 223.—T. S. leaf: *e*, upper epidermis; *g*, crystalline granules; *p*, palisade cells. (× 500).

Fig. 224.—T. S. leaf: *l*, lower epidermis; *s*, spongy tissue. (× 500).

Fig. 225.—T. S. leaf, showing the festoons of cells around a vein. (× 240).

Fig. 226.—T. S. leaf, showing the festoon around a longitudinally cut vein. (× 240).

Fig. 227.—T. S. stem: mesophytic form. (× 500).

Fig. 228.—Leaf: lower epidermis of the mesophytic form. (× 240).

Fig. 229.—T. S. leaf: mesophytic form. (× 240).

Celosia argentea, Linn.

Fig. 230.—Photograph, showing the halophytic forms.

Fig. 231.—Photograph, showing the mesophytic forms.

Fig. 232.—Stem: epidermis from the ridge. (× 240).

Fig. 233.—   do.   do.   from the groove. (× 240).

Fig. 234.—T. S. young stem, showing palisade-like cells of the cortex. (× 500).

Fig. 235.—T. S. stem: *c*, collenchyma; *ch*, chlorenchyma, (× 240).

Fig. 236.—T. S. petiole: c, cortical cells. (× 500).

Fig. 237.—T. S. petiole (semi-diagrammatic): k, crystal cells; v, vascular bundles; ch, chlorenchyma. (× 41).

Fig. 238.—Leaf: upper epidermis. (× 240).

Fig. 239.—Leaf: lower epidermis. (× 240).

Fig. 240.—T. S. leaf: e, upper epidermis; p. palisade cells. (× 240).

Fig. 241.—T. S. leaf: l, lower epidermis; s, spongy tissue. (× 240).

Fig. 242.—T. S. leaf, showing the sheath around a vein: p, palisade cells; k, crystal cells; v, vein. (× 240).

Fig. 243.—T. S. old leaf: e, upper epidermis; p, palisade cells. (× 240).

Fig. 244.—T. S. old leaf: l, lower epidermis; s, spongy tissue. (× 240).

Fig. 245.—T. S. petiole of the mesophytic form: ch, chlorenchyma. (× 41).

Fig. 246.—Leaf: lower epidermis of the mesophytic form. (× 240).

Fig. 247.—T. S. leaf: mesophytic form. (× 240).

Sesuvium portulacastrum Linn.

Fig. 248.—Photograph, showing the halophytic form.

Fig. 249.—T. S. stem, showing the outer cortex. (× 500).

Fig. 250.—T. S. stem, showing the inner cortex. (× 240).

Fig. 251.—Leaf: upper epidermis. (× 240).

Fig. 252.—T. S. leaf: e, upper epidermis with a stoma. (× 500).

Fig. 253.—T. S. leaf: e, upper epidermis; p, palisade cells. (× 240).

Fig. 254.—T. S. leaf: p, palisade cells; a, aqueous tissue; k, crystal cell. (× 240).

Fig. 255.—T. S. root, showing the lacunar cortex with starch. (× 240).

Fig. 256.—T. S. root, showing the anomalous growth. Explanation in the text. (× 30).

Suaeda nudiflora, Moq.

Fig. 257.—Stem: epidermis. (× 500).

Fig. 258.—T. S. stem: g, guard cell of the abnormally placed stoma. (× 500).

Fig. 259.—T. S. stem, showing the cortex. (× 500).

Fig. 260.—T. S. stem: p, phellogen; h, hard bast. (× 240).

Fig. 261.—Leaf: upper epidermis. (× 240).

Fig. 262.—Leaf: lower epidermis. (× 240).

Fig. 263.—Leaf: section parallel to the main vein, showing a stoma. (× 240).

Fig. 264.—Photomicrograph: T. S. leaf. (× 82).

Fig. 265.—T. S. leaf: *p*, palisade cells; *c*, collecting cells; *a*, aqueous tissue; *g*, guard cell. (× 240).

Fig. 266.—T. S. leaf. (× 51).

Fig. 267.—Photograph, showing the thick old leaves.

Fig. 268.—Photomicrograph: T. S. root. (× 30).

Arthrocnemum indicum, Moq.

Fig. 269.—Stem: epidermis. (× 500).

Fig. 270.—Tangential L. S. stem, showing a stoma. (× 500).

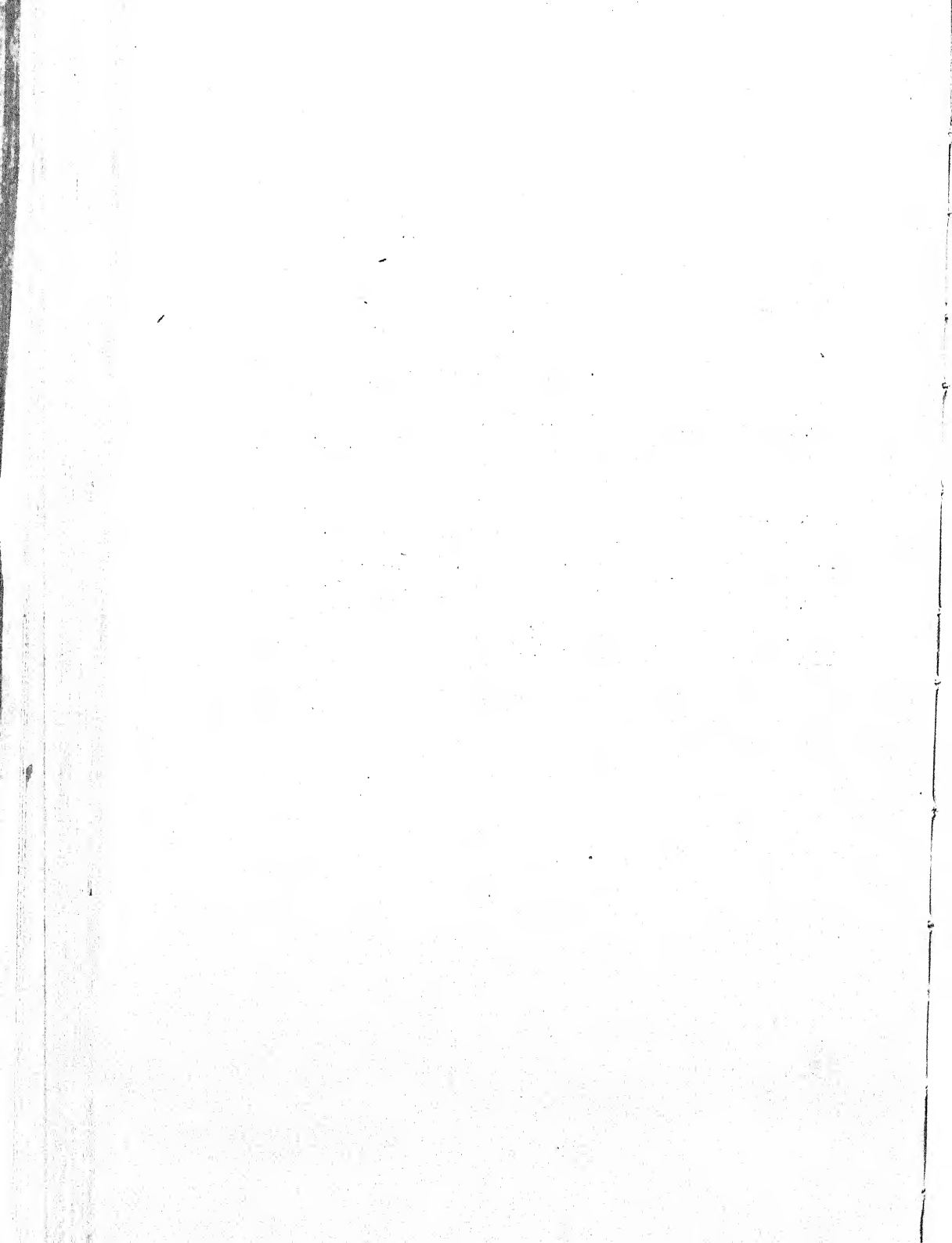Fig. 271.—Photomicrograph: T. S. stem. (× 60).

Fig. 272.—T. S. stem: *p*, palisade cells; *a*, aqueous tissue; *g*, guard cell; *v*, vascular bundle. (× 150).

Fig. 273.—Photomicrograph: T. S. root. (× 180).

Fig. 274.—T. S. stem of the mesophytic form: *p*, palisade cells; *v*, vascular bundle; *a*, aqueous tissue. (× 150).

Fig. 275.—Photomicrograph: T. S. stem of the mesophytic form. (× 60).

Plate II.

198

199

201

205

200

202

203A

203B

204

206

207A

207B

208

213

210

211

209

212

214A

214B

216

215

217

219B

219A

218

220

221

222

223

253

254

256

A

B

252

256 C

256 D

255

Plate IX.

257

258

259

260

261

262

263

265

269

270

266

272

274
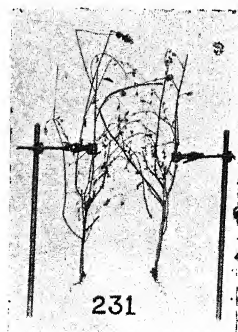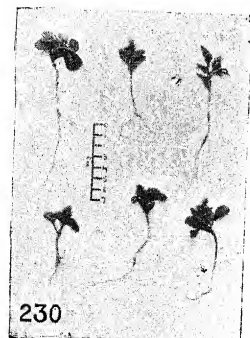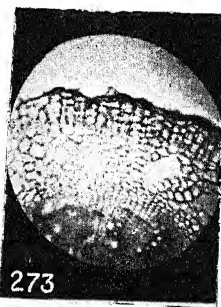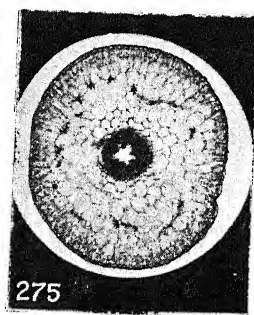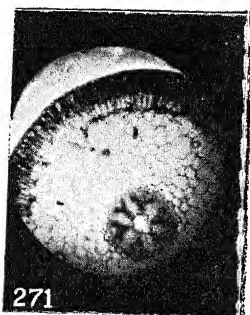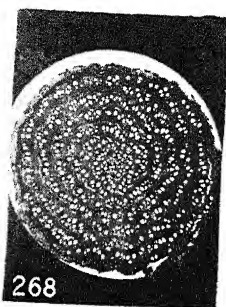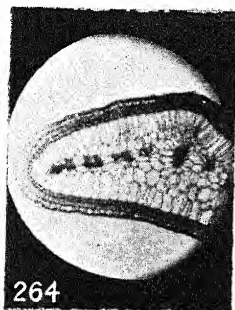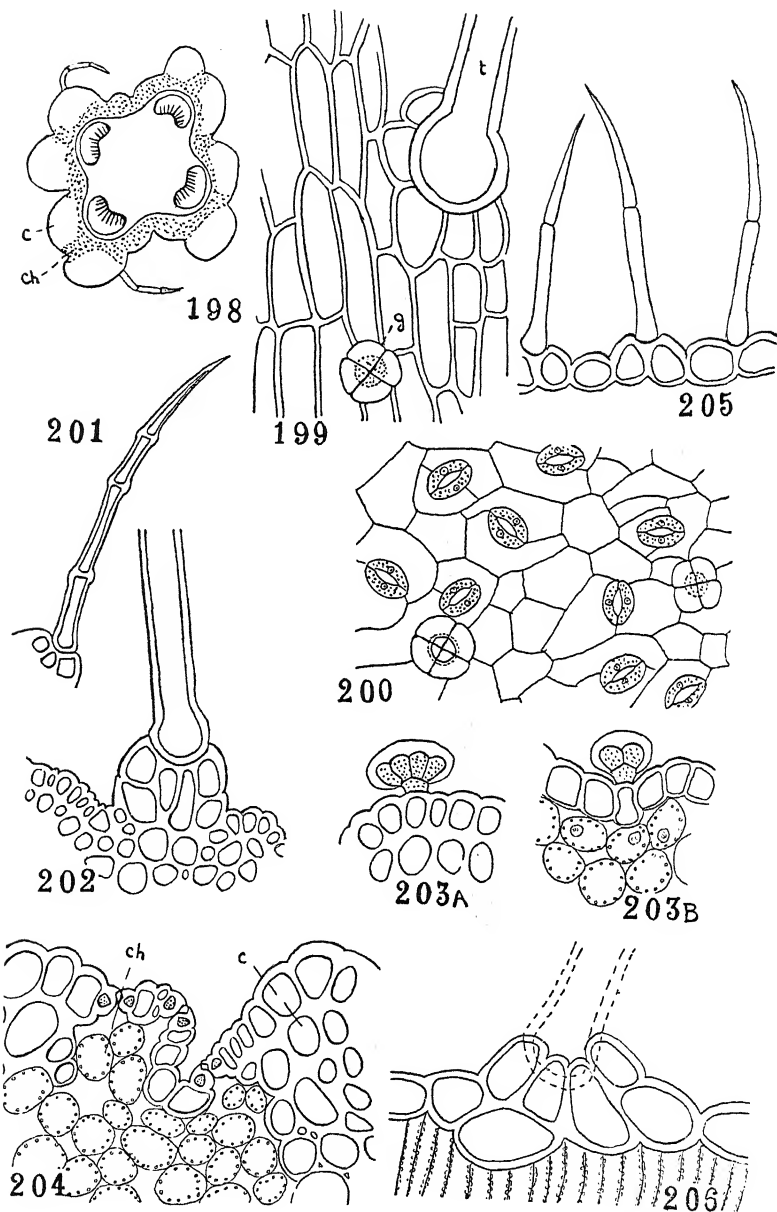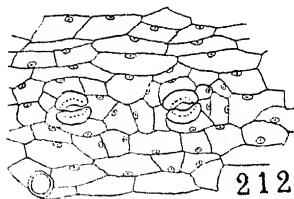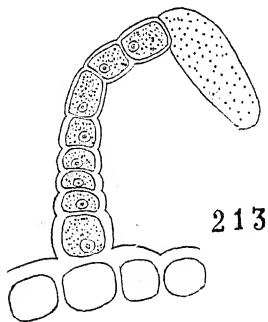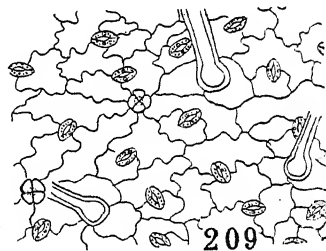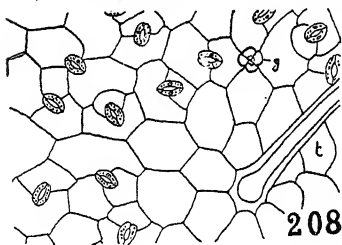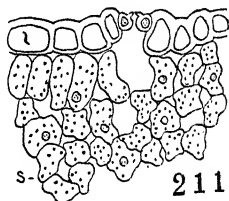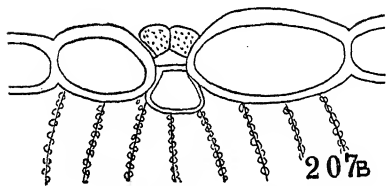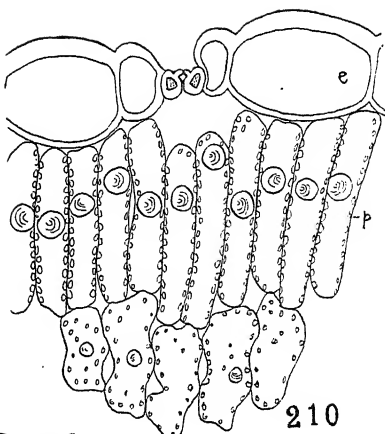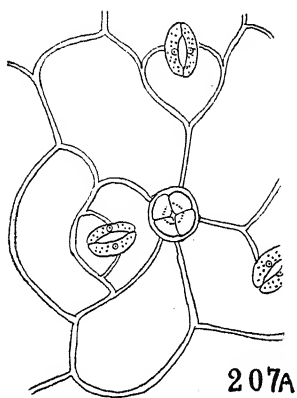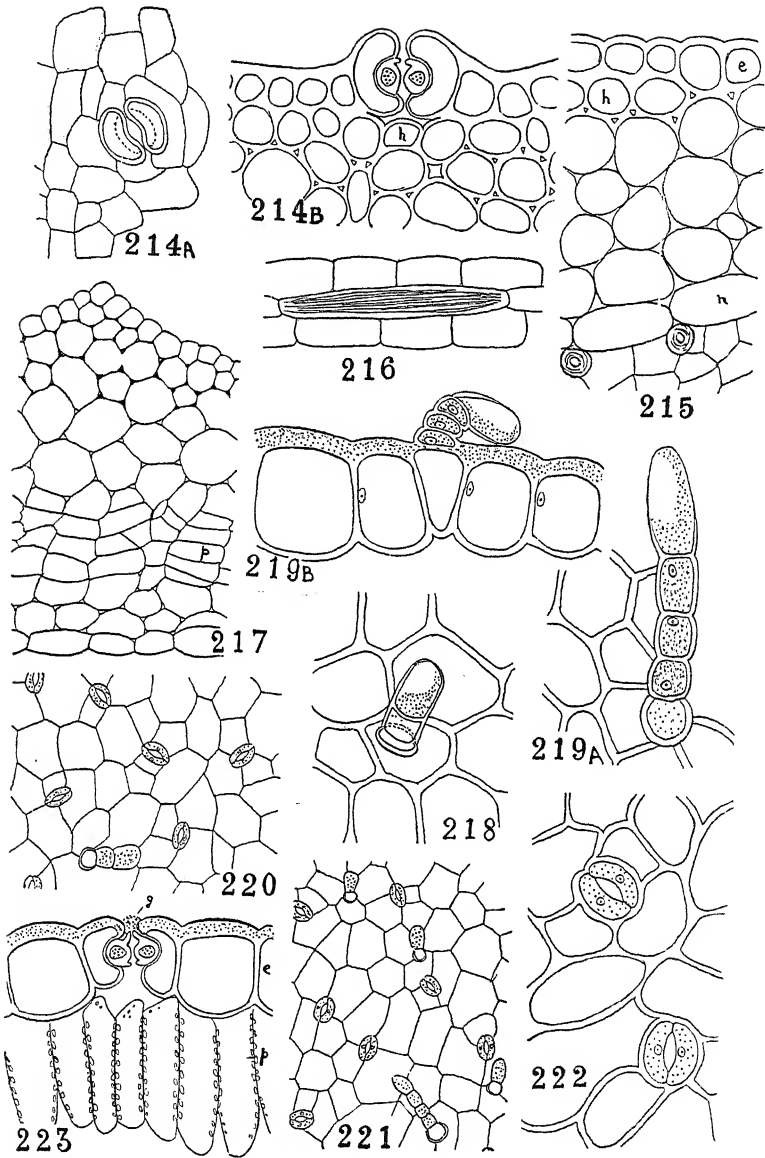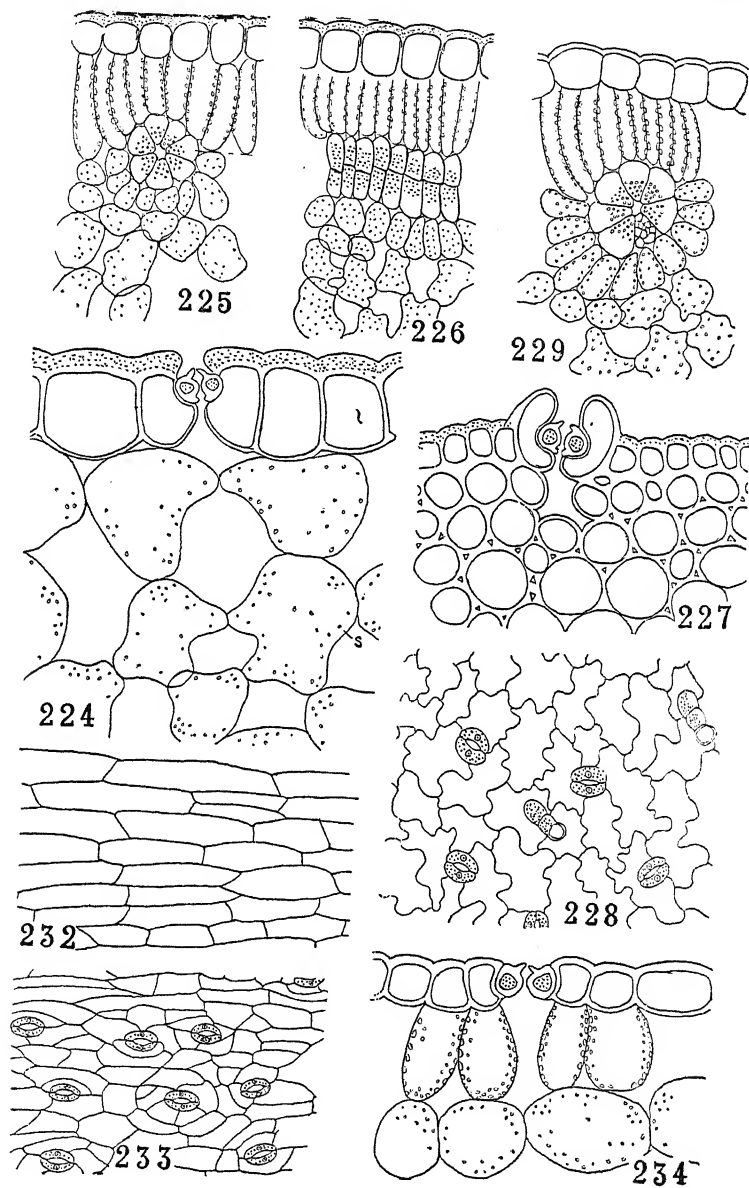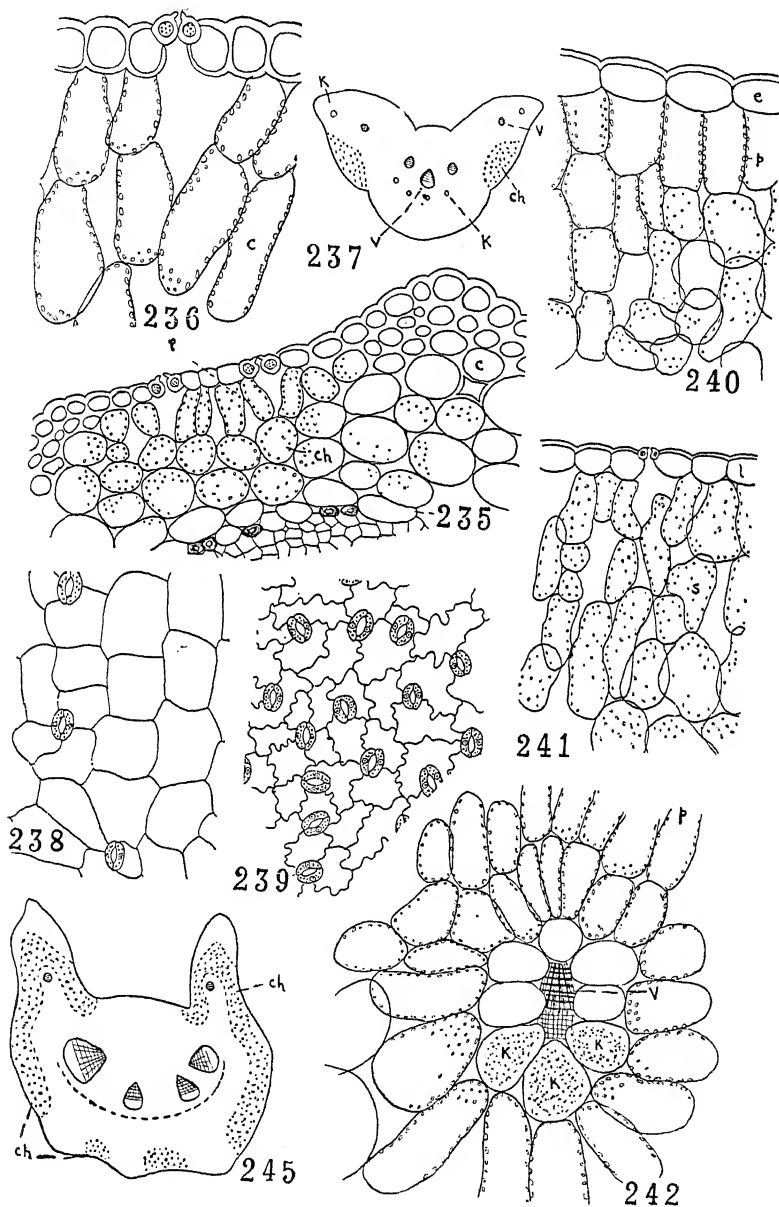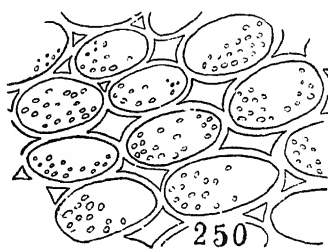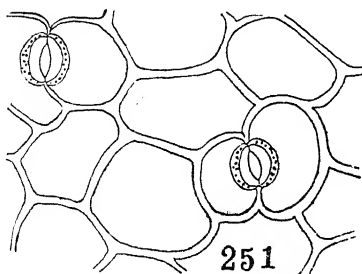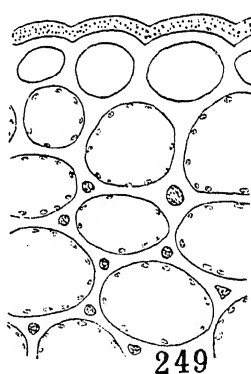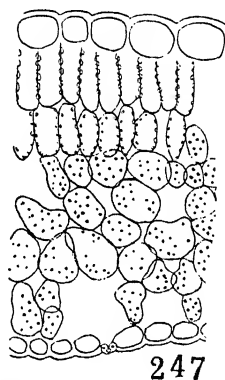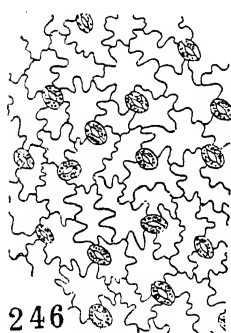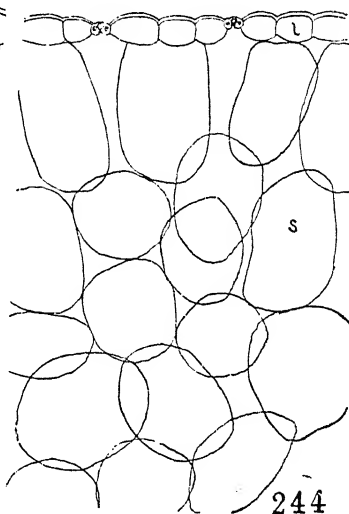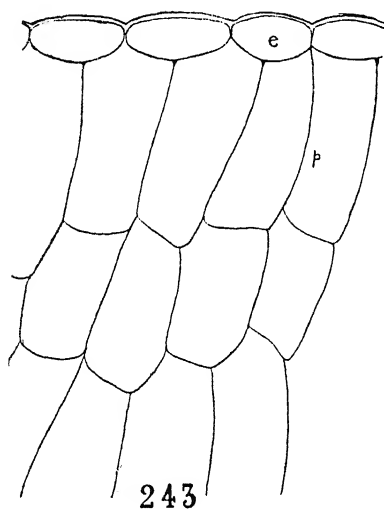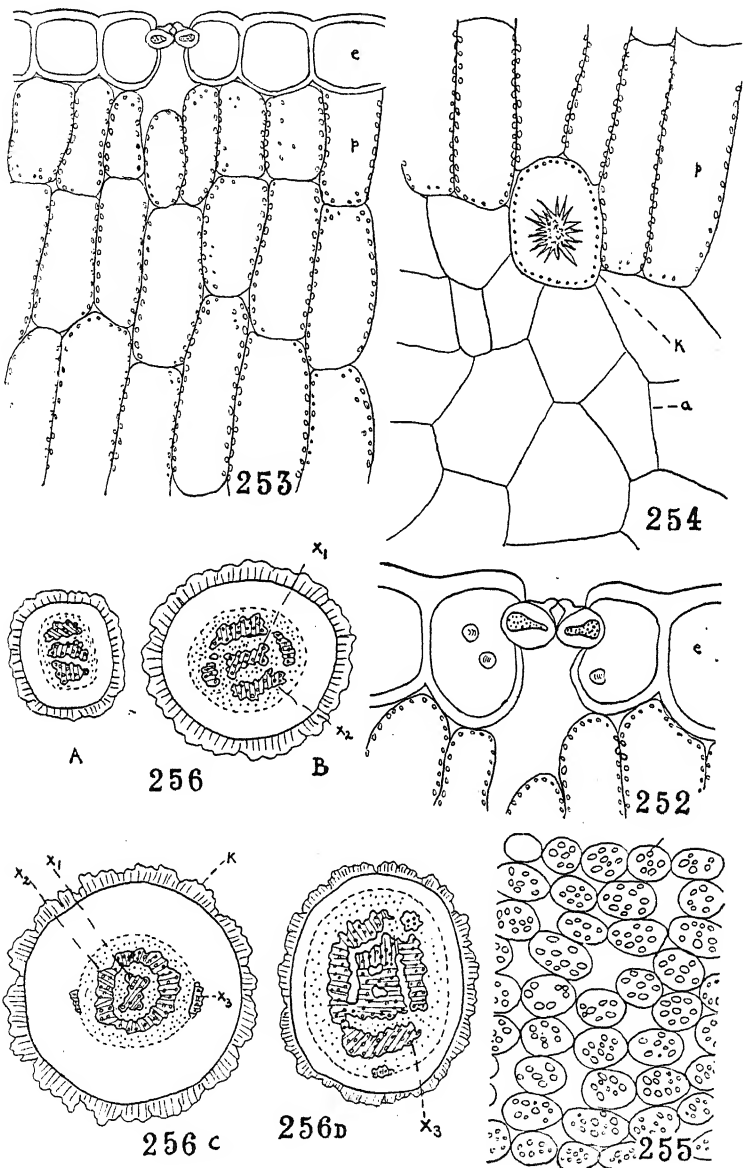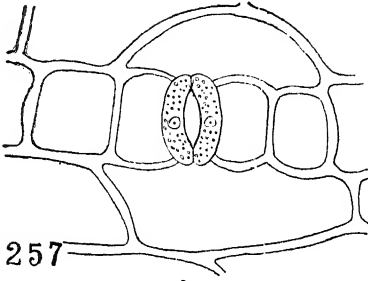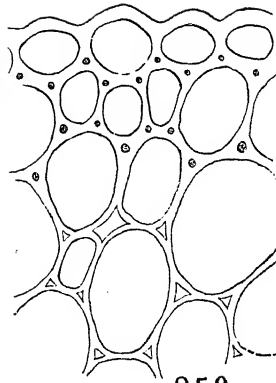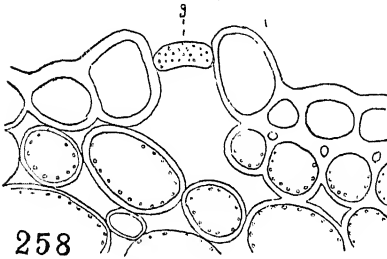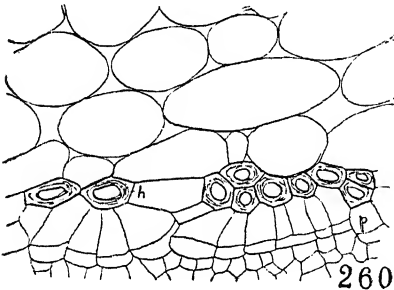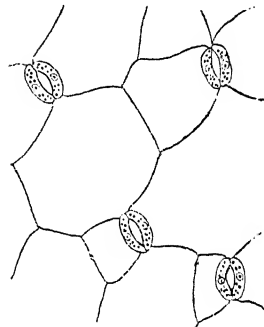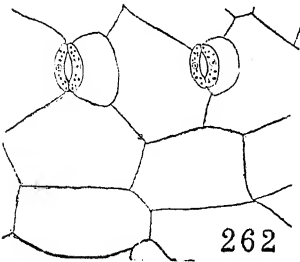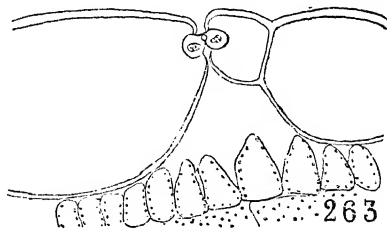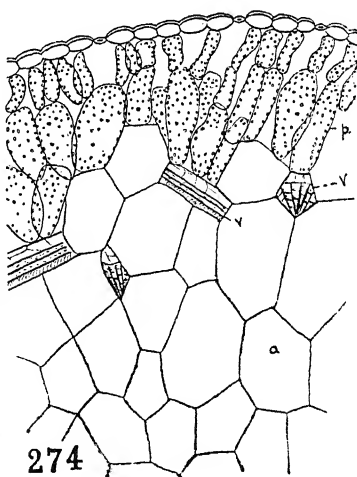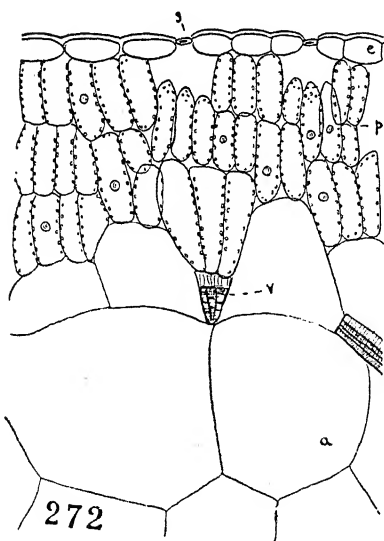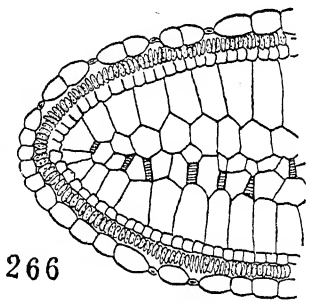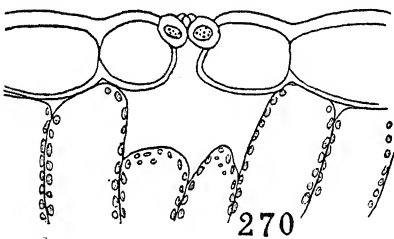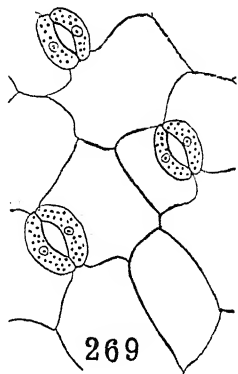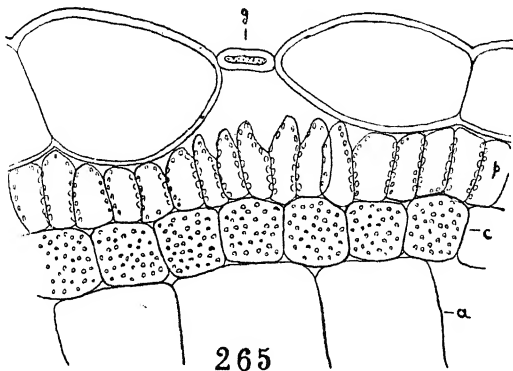
SOME ABNORMAL FLOWERS OF *ARGEMONE MEXICANA*, LINN.

BY

A. C. JOSHI,

Benares Hindu University, Benares.

With one plate and nine figures in the text.

Introduction.

Three plants of *Argemone mexicana* Linn. (Papaveraceæ), bearing flowers very different from the normal, were collected by the writer in the first week of April, 1931, from a field near Hoshiar Pur in the Sub-Himalayan tracts of Punjab. The present paper describes the external morphology of these abnormal flowers and the structure of their stamens and the ovules.

The usual methods were employed in the investigation. Twigs from two abnormal plants were removed and taken to the Botanical Laboratory of Government College, Lahore, where these were photographed. After this they were preserved in rectified spirit. About three days elapsed between the plucking of the twigs and their final preservation in alcohol. During this period they were kept in a bottle of water as shown in Fig. 10, but even then they had begun to wither at the time of fixing. The third plant was left at its place undisturbed to see if it may set some seed and leave some offsprings behind. For this purpose the place was again visited by my brother in May 1932, but no such plants were found. The preserved material was finally removed to the Benares Hindu University, in the Botany Department of which, the rest of the investigation has been carried out.

External Morphology.

The normal plants of *Argemone mexicana* are erect glaucous prickly herbs, quite robust in their construction, often almost woody below and reach a height of one and a half to four feet. Branching is not very abundant and often the stem remains throughout unbranched and simple. The leaves are upto eight inches long, sessile, semi-amplexicaul, sinuate-pinnatifid and white-spotted. The flowers are large and showy, about 2 inches in diameter, sessile or subsessile, mostly yellow, but Duthie (6) mentions flowers with white petals also. They conform to the

5

floral formula K_3 C_3 $A\infty$ G (4-6).   The sepals are green, horned at the top, bristle-pointed and fall off with the opening of the flowers, i.e., they are caducous, as in the whole family Papaveraceæ. Petals are deciduous and wither away after pollination. Stamens are indefinite in number, hypogynous. They possess slender filaments with a weak vascular strand and anthers which are nearly 3 times as long as broad, 4-locular, and dehisce longitudinally (fig. 1a). Ovary is superior, elliptic or oblong, bristly, 1-celled, with numerous ovules on 4–6 parietal placentas. Style is very short, nearly absent and the 3–6 lobed stigma is nearly sessile. Capsule is $1–1\frac{1}{2}$ inches long, prickly and opens at the top by short valves.

The abnormal plants were quite similar to the normal ones in habit and vegetative characters. This is quite clear from fig. 10, which is a photograph of these plants. The following differences, however, were seen in the flowers :—

(1) The sepals did not fall off on the opening of the flowers. These were found to persist on the torus even up to the last stages of the flower.
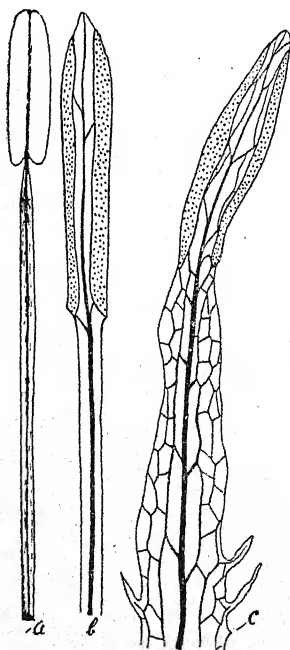
Fig. 1. *Argemone mexicana. a,* stamen from a normal flower; *b* and *c,* stamens from two abnormal flowers, showing phyllody to a varying extent. × about 10.

(2) The petals were not yellow. Many were green and others showed various gradations between green and yellow. Further, like the sepals they were also found to be persistent even in the oldest flowers.

Both of these features can be seen from fig. 10.

(3) Stamens were found to show a tendency towards phyllody though there were none which had become completely foliaceous. The stamens of the different flowers were not of the same form. In some cases the process had shown itself but slightly; in other cases it was more pronounced. Two cases are sketched in figs. 1*b* and 1*c*, as illustrating the amount of variation. The first differs from the normal stamen, shown in fig. 1*a*, only in some increase in the breadth of the filament and connective, an increase in the size of the vascular strand and its branching in the region of the connective. In the stamen sketched in figure *1c, the* filament and the connective are both very much enlarged and the vascular tissue shows copious branching and anastomosing, so that a well developed reticulum of veins is formed both in the region of the connective and the filament. Further, the filament shows bristly outgrowths such as are characteristic of the vegetative parts.

In all the abnormal flowers, the anthers of the stamens never opened and the stamens never withered and were found to be present even in the oldest flowers.
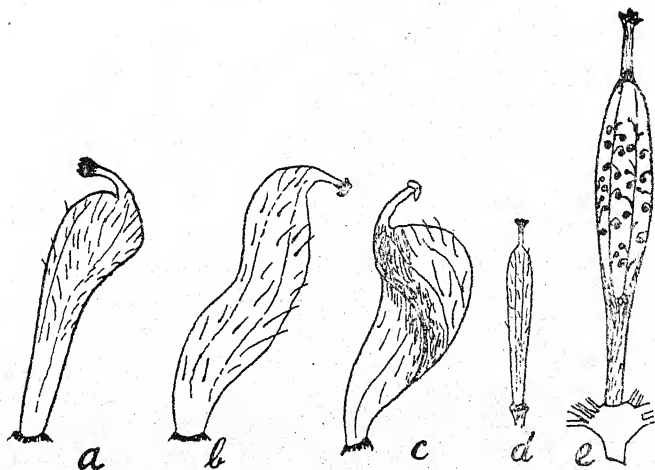
Fig. 2. *Argemone mexicana. a-d*, pistils from different abnormal flowers, *a-c* from old flowers and *d* from a flower-bud; *e*, V.S. of the pistil sketched in *d. a-d* about natural size; *e*, thrice the natural size.

(4) The gynæcium showed even more characteristic differences. In many cases there was a short stalk, gynophore, developed below the ovary (figs. 2, *d* and *e*), a feature characteristic of the Capparidaceæ. There was always a well-developed style (figs. 2, *a-e and* fig. 10). The ovary usually showed, especially in the older flowers, various types of curves which gave it a zygomorphic symmetry (figs. 2, *a-c,* and fig. 10). Sections of the ovaries (fig. 2*e*) showed that the ovules inside were less numerous, longer-stalked and more stout as compared with those of the normal flowers and the curves of the ovary in the older flowers were due to the devlopment of the ovules in some parts of the ovary and their absence at other places.

The ovules did not mature into seeds. No fruit was formed and the ovary never opened. The plants thus died, at the end of the growing season without leaving behind anything of the nature of reproductive bodies.

Structure of the Stamen.

a. Filament. The structure of the filaments of normal and abnormal stamens is shown in text-figure 3 (*a-d*). Figs. 3*a* and 3*b* represent transverse sections of filaments of normal stamens. The one represented in 3*a* shows the usual structure. There is epidermis on the outside without any stomata, parenchymatous mesophyll without any green chloroplasts and a single collateral vascular strand in the centre with the xylem directed towards the centre of the flower, i.e., on the ventral side and phloem on the dorsal side. Fig. 3*b*, shows that at some places,—this is usually just below the anther where the filament is very narrow (see fig. 1*a*)—the filaments of normal stamens are devoid of any vascular tissue. Figure 3*c* shows a transverse section of a filament from a type of stamen sketched in fig. 1*b*. It differs from the filament of a normal stamen only in its larger size, slight flattening and in the presence of a better developed vascular strand. The transverse section of the filament of the stamen sketched in figures 1*c* is shown in figure 3*d*. Not only is this filament larger, but it is more flattened and leaf-like. There is differentiation of midrib and wings and mesophyll of the latter contains chloroplasts in abundance. Stomata are present both on the upper and the lower surface in the region of the wings and their guard-cells are slightly sunken below the surface. The vascular tissue is very well-developed. The main bundle is present in the centre of the midrib and a number of its branches are seen ramifying in the wings. These

features in addition to those described previously are other signs
of a tendency towards phyllody in the andrœcium of these abnor-
mal flowers.
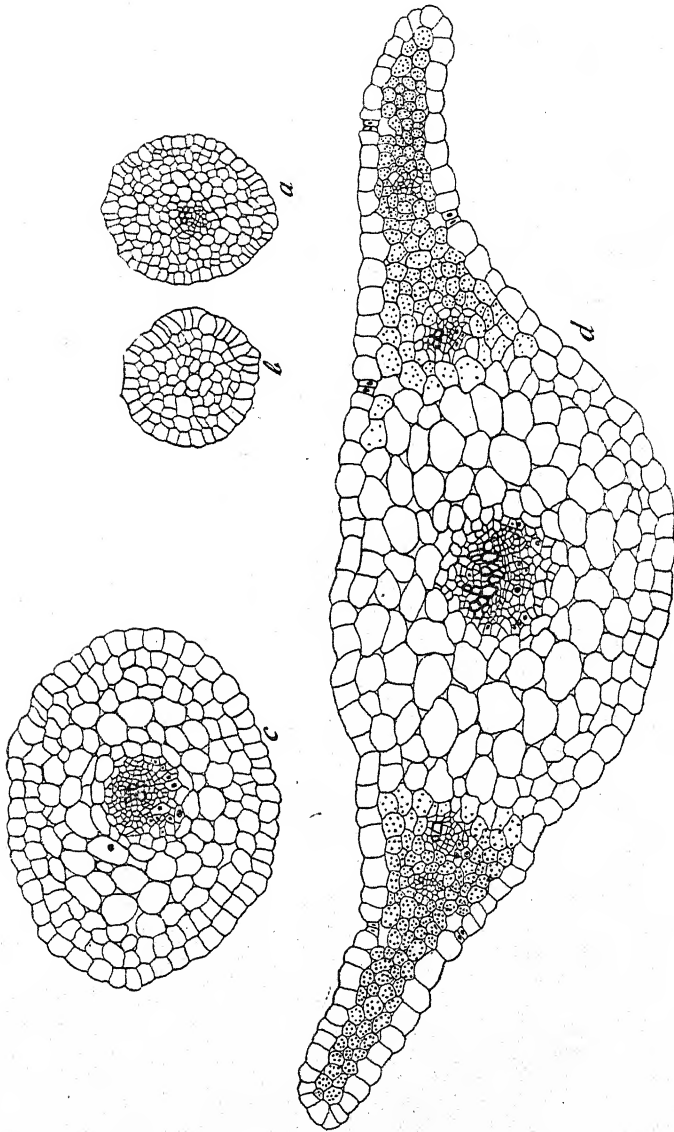
Fig. 3. *Argemone mexicana.* Transverse sections of the filaments from
different stamens; *a* and *b*, of a stamen from normal flowers,—in *b* there is
no vascular bundle; *c*, T.S. of the filament from a type of stamen sketched in
fig. 1*b*; *c*, T.S. of the filament from a type of stamen sketched in fig. 1*c*.
× 115.

b. Anther. The structure of the anthers of the normal and the abnormal stamens of *Argemone mexicana* is shown in text-figure 4 (*a-c*). Fig. 4*a* is a transverse section of the anther of a
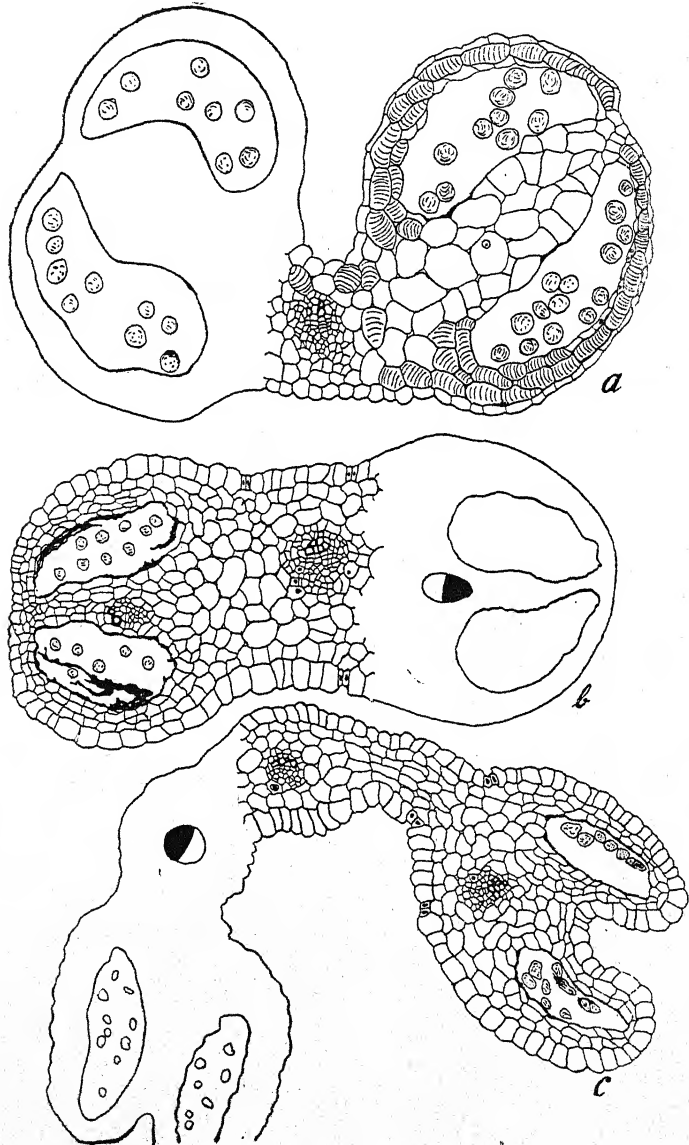
Fig. 4. *Argemone mexicana.* Transverse sections of the anthers from different stamens; *a* of a normal stamen; *b*, from the type of stamen sketched in fig. 1*b*; *c* from a type of stamen sketched in fig. 1*c*. × 115.

normal stamen, 4*b* of the anther from a stamen of the type sketch-
ed in figure 1*b* and 4*c* of an anther from a stamen of the type
sketched in figure 1*c*. The anther sketched in fig. 4*a* is from an
unopened bud and it has not dehisced as yet. It shows a short
connective, 4 pollen sacs with a wall about 3 cells thick. There
is a well developed endothecial layer with its characteristic
fibrously banded cells. There is single unbranched vascular strand
in the region of the connective and there are no stomata. The two
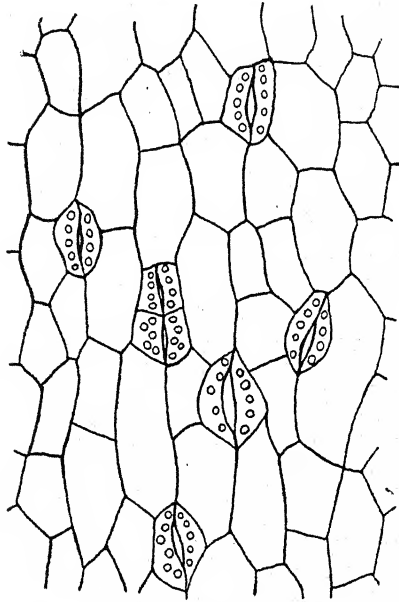
Fig. 5. *Argemone mexicana*. Epidermis of the connective from a type
of stamen shown in fig. 1*b* in surface view, showing simple and double stomata.
× 325.
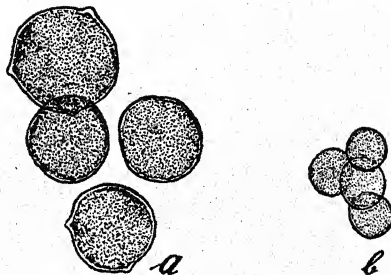
Fig. 6. *Argemone mexicana*. Pollen grains; *a*, from normal flowers; *b*,
from abnormal flowers. × 225.

anther-halves are bent inwards and the whole structure is in no way different from that usual for introrse, longitudinally dehiscing anthers. The pollen grains are nearly 50 microns in diameter.

In the anthers of the abnormal stamens the connectives are much larger, especially in the type sketched in figure 4c, a feature which was also visible externally. There are stomata in the epidermis of the connective both on the dorsal and the ventral sides. These are of the same form as are found on the filament. Their surface view is shown in figure 5. They are usually simple, but sometimes double stomata are also found. There is no differentiated endothecial layer with fibrous cells. This explains the loss of dehiscing power in the anthers. The vascular tissue also in the abnormal anthers is better developed. Besides the median bundle, a transverse section usually shows two more bundles, one on each side, situated in between and near the pollen-sacs, the anther-halves of the normal stamens. The anthers open outwards (fig. 4c), in a direction quite opposite to that followed by the anther-halves of the normal stamens. The anthers of the stamens of the type sketched in figure 1b are nearly straight (fig. 4b) and thus present a condition intermediate between the first two types. The pollen grains of the abnormal stamens were found to vary between 12–20 microns in diameter and thus these were only about one-fourth to one-third in size as compared with the pollen grains from the normal stamens (text-fig. 6). Further the contents of these pollen grains were much less dense and they appeared to be more or less empty.

Structure of the Ovule.

The ovules from the abnormal flowers of *Argemone mexicana* were found to differ from the normal ovules in the structure of almost every part, namely, the integuments, the nucellus, the embryo-sac and the vascular supply. A sketch of a normal ovule has already been published by the writer in an earlier paper (13). It was found that in normal ovules the integuments are fused with each other and the inner one with the nucellus at maturity. The apex of the nucellus is nearly blunt and there is a short micropyle. The embryo-sac is of the normal 8-nucleate type and, when mature, eats up nearly the whole of the nucellus which then forms only a very small layer around it. There are neither chloroplasts, nor stomata, either in the integuments or in the nucellus. The vascular supply of the ovule ends at the junction of the nucellus and the stalk of the ovule in the region of the chalaza.

In the ovules from the abnormal flowers it was found that the integuments were perfectly free both from each other and the nucellus. They were much better developed than in the ovules from ordinary flowers of the species and formed a greatly elongated micropyle (text-fig. 7a). The apex of the nucellus was also acute and somewhat prolonged into the elongated micropyle. The structure of the integuments further was very characteristic.
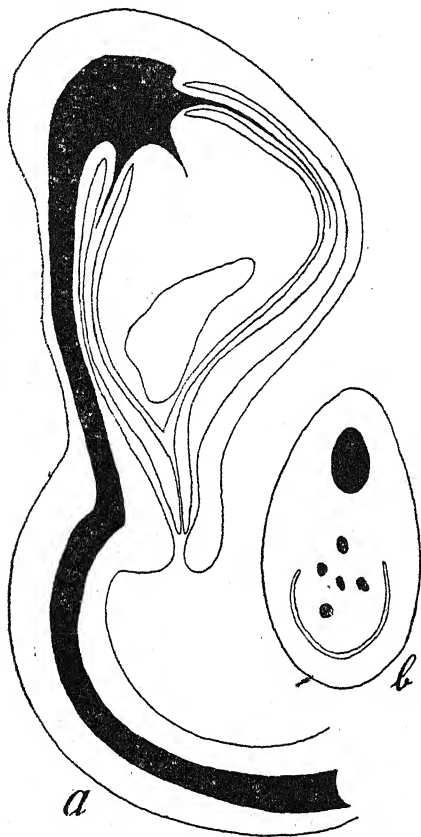
Fig. 7. *Argemone mexicana*. *a,* longitudinal section of an ovule from an abnormal flower, showing free integuments, long micropyle, a well-developed vascular strand which in the region of the chalaza sends out similar branches into the base of the nucellus and the inner integument; the embryosac is represented by an empty space; *b,* a rather oblique section through a beginning to get free from the nucellus, showing the vascular bundle of the funiculus and the branching of that bundle in the chalaza into 5 strands, one of which is more or less in the centre and 4 around that more or less in a ring. Vascular tissue is represented black. × 65.

Each had an inner and outer epidermal layer consisting of large cells without any chloroplasts and 1–5 layers of the mesophyll between the two epidermal layers. The cells of the mesophyll layers were abundantly provided with chloroplasts. The outer epidermis of the integuments had a few stomata, but the inner epidermal layers of both the integuments were abundantly provided with these and the two integuments of the ovule were thus perfectly equipped for work of carbon-assimilation (fig. 8). In fact they were perfectly leaf-like in structure. The structure of the stomata found on the ovules was quite similar to those found on the stamens.

. The embryosac of all the abnormal ovules studied by the writer was found to have completely degenerated and left behind only an irregular cavity in the nucellus (fig. 7a). The latter was well developed and it had not been destroyed by the embryosac to the same extent as in the normal ovules. Further its outer layers just beneath the epidermis were well provided with chloroplasts and in the living state it must have been green and able to do some assimilatory work (fig. 8).
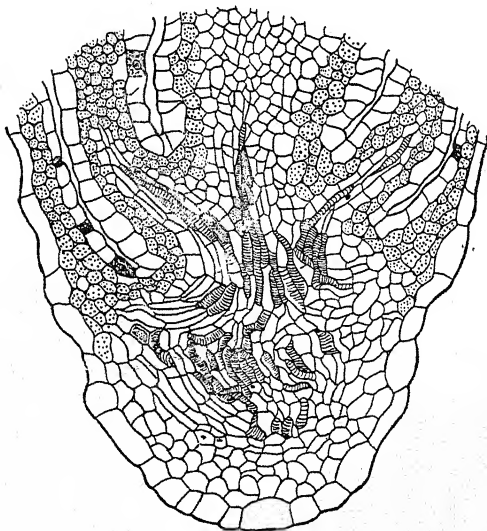
Fig. 8. *Argemone mexicana*. Chalazal portion of the ovule shown in *fig. 7a*, showing the branching of the vascular bundle and the going of these branches into the base of the nucellus and the inner integument. The mesophyll of the integuments and the peripheral part of nucellus shows the presence of chloroplasts. There are stomata in the integuments on the inner side. × 115.

Very important differences were seen in the vascular supply of the abnormal ovules, as compared with that of the normal ones. The vascular bundle of these ovules was much better developed (fig. 7a) and it did not terminate in the region of the chalaza (figs. 7a, 7b and 8). On the other hand, at this point it divided into 5 or 6 small strands, one situated in the centre and the others forming an irregular ring around it (fig. 7b). The central strand was found to supply the base of the nucellus and the outer ones the inner integument (figs. 7a and 8). They ran into these tissues for a short distance and then ended blindly, but in one case one of the integumentary strands was found to run nearly upto half the length of the ovule.

Discussion.

The nature and the cause of the above abnormalities. From the description of the abnormal plants given above, it is quite clear that even though their vegetative parts were quite similar to those of the normal plants, they showed many important differences in the organisation of their flowers. Further these differences were to be found in every part of the flower, the calyx, the corolla, the andrœcium and the gynæcium, having been all affected by the change, which had collectively led to the development of these peculiar flowers. What the nature of this change or abnormality is, is fairly clear, though it is only possible to guess at the cause. The fact that both the sepals and the petals had become persistent and the latter also virescent to a varying extent, shows that both the parts were becoming leaflike and were affected by a change of a phylloid nature. The same is true of the andrœcium and the gynæcium. The broadening of the filaments and the connectives of the stamens, the development of bristle-like outgrowths on the filaments, of stomata and chlorenchymatous tissue both in the filaments and the connectives, the formation of the stomata in the integuments of the ovules and the development of chlorophyll both in the mesophyll of the integuments and in the outer portions of the nucellus are all characters of the green leaf. The loss of the fibrously banded endothecial layer in the anthers also makes the stamens more comparable with the leaves. Only a few differences like the development of the style between the stigma and the ovary and of the small gynophore below the ovary in some cases seem to be unconnected with characters of the green leaves. Taking all these facts into consideration it appears to be quite clear that these abnormal flowers of *Argemone mexicana* were af-

fected by a change of the nature of partial phyllody. It had made the two outer whorls of floral leaves, always more leaflike as compared with the two inner whorls, still more foliaceous, while it had added a few characters of the green leaves to the androecium and the gynæcium. Correlated with this change there have appeared a few variations, mentioned above, of an independent nature, some of which, like the development of the style, are probably of the nature of reversions.*

As regards the cause of the development of these abnormal flowers, as said above, it is only possible to guess. The plants as they were found growing in the field appeared to be quite healthy. After they had been brought to the laboratory, all their parts were subjected to a thorough microscopic examination to see if they may not be affected by some fungus or insect pest, but no such was found on them. The probable cause of these abnormalities then perhaps lies in the very constitution of the plants and is of an internal nature. This is completely supported by the fact that all the flowers of the plants were affected by the change. What this constitutional change was, however, is a question belonging to the sphere of Plant Genetics and Cytology. In the absence of such knowledge one can only compare his observations with what is known to occur in other plants. The pollen from the abnormal plants was found to be much smaller in size and more or less empty as compared with the pollen from normal plants. It is commonly known that abnormal pollen is sterile. The embryo-sac in the ovules of the abnormal flowers also degenerated and never produced any embryo. This sterility of the spores, both of the microspores and the megaspores has been recognised as a characteristic of hybrids even from such a time as that of Dutrochet (7) and Gaertner (9) among recent botanists by Rosenberg (19), Juel (15), Tischler (23), Dorsey (5), Jeffrey (14), Hoar (11), Cole (2) and many others. With the exception of DeVries (4), this view seems to be generally accepted and it would lead us to the conclusion that the abnormal flowers of *Aregmone mexicana* described in the present paper,

---

* This statement is based on the fact that a style is present between the ovary and the stigma in the majority of flowering plants. It is present in the other families of the Rhoeadales, like Capparidaceae and Cruciferæ and even in several other Papaveraceæ. There can be little doubt that it was also present in the ancestors of *Argemone, Papaver* and other genera of Papaveraceæ with sessile stigmas. The development of style thus in abnormal cases in *Argemone* would be a reversion.

also owe their origin to hybridisation. Such a conclusion seems to be well supported from the facts that the petals and the stamens of the abnormal flowers were not all alike,—they showed various gradations in virescence and phyllody, just as would sometimes happen on segregation after a cross, that hybridisation does produce new forms,—this can be seen from the work of a number of authors like that of Lotsy (18) on *Antirrhinum* and *Lychnis*—and that hybridisation does occur in *Argemone*. The last feature has been seen by the writer by an examination of the pollen of a number of plants growing at Benares. It has been seen that in several cases about 50% of the pollen from an anther is sterile (fig. 9), even in flowers which are in other respects perfectly normal. What type of cross it was that had given rise to these abnormal flowers of *Argemone* is again a question relating to the science of Plant Genetics and nothing can be said about it.
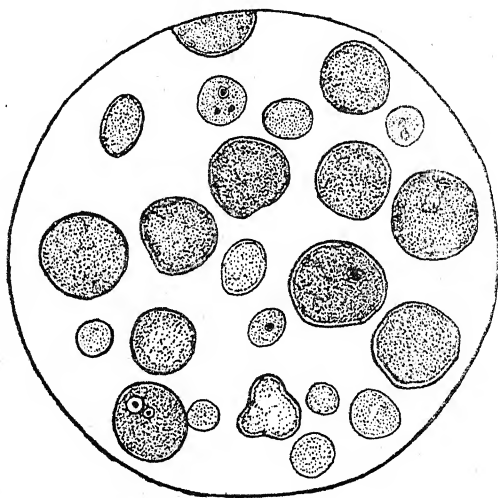
Fig. 9. *Argemone mexicana.* Pollen from a normal plant, selected at random, showing a large number of sterile grains. × 230.

The function of the endothecium. It is generally recognised that the function of the fibrous endothecial layers in the anthers of the flowering plants is to help in dehiscence (Coulter and Chamberlain, 3). The anatomy of these abnormal flowers of *Argemone*, so far as it goes, gives full support to this generally accepted conclusion. The anthers of the normal stamens of *Argemone* are provided with a well-developed fibrous endothecium, and they always dehisce. The anthers of the abnormal flowers are devoid

of such a differentiated endothecium and never open, cleary showing thereby the use of this endothecial layer.

Phylogenetic considerations. Whether evidence from the anatomy and morphology of abnormal plants or teratological specimens should be used in discussions pertaining to the phylogeny of plants is an old much discussed question. Many writers have used such an evidence before and consider it to be of utmost importance. Others consider it to be more or less useless. A discussion of the subject is given by Worsdell (25) in the introductory part of his "Principles of Plant Teratology". The writer does not want to enter into any such discussion here, but to simply state a few points which seem to emerge from the present study and appear to be of some phylogenetic importance.

The family Papaveraceæ to which *Argemone* belongs is considered by every modern taxonomist to be related to such families as Cruciferæ, Capparidaceæ, etc., and these are together placed in the order Rhoeadales (Engler and Gilg (8), Wettstein (24), Hutchinson (12), etc.). One of the points in which it differs from the rest is the absence of the style in many genera. The presence of the style in the abnormal flowers of *Argemone* described here, which seems to be a sort of reversion, makes it more comparable with the other families. The same thing may be said about the development of the gynophore below the ovary in some cases. The presence of a gynophore is one of the characteristic features of the family Capparidaceæ and its development in the abnormal flowers of *Argemone*, emphasizes the relation of the Papaveraceæ to the Capparidaceæ.

Another point of interest in this connection is the great development of the vascular system in the ovules of the abnormal plants. In the normal plants of *Argemone* the vascular supply of the ovule ends in the region of the chalaza, but in the abnormal plants the vascular bundle at this point branches out and the various branches pass into the inner integument and one, the central one, to the base of the nucellus. A well-developed vascular supply to the integuments and the base of the nucellus was a feature of the pteridospermous ovules. It is found in a few angiosperms at the present time, as for example, in *Myrica* (Kershaw, 16), Juglandaceæ and some other Amentiferæ (Kershaw, 17 and Benson and Welsford, 1), some Thymelæaceæ (Guerin, 10). Further evidence is also accumulating that the modern angiosperms are derived from some pteridospermous stock (Thomas, 21, 22).

Seward (20) regards this view as the most acceptable one at the present time. The presence of pteridospermous features in the ovules of *Argemone* in abnormal cases, which show some features of reversion, lends, so far as it goes, its support to such a view.

Summary.

Some abnormal flowers of *Argemone mexicana* L. are described. Their main features are the persistent calyx and corolla, the latter also showing virescence to a varying extent, broadening of the filaments and connectives of the stamens, development of bristly outgrowths on the former and of stomata and chlorenchymatous tissue in both, loss of the fibrous endothecium in the anthers, freedom of the integuments of the ovule both from each other and the nucellus, long microphyle, development of stomata on the integuments and of chlorophyll both in the integuments and in the outer layers of nucellus, a strong development of the vascular tissue both of the stamens and the ovules, the branching of the vascular bundles in the chalaza of the ovules, and the running of these branches into the inner integument and base of the nucellus.

The nature, cause and phylogenetic significance of these abnormalities is discussed. They appear to be of the nature of partial phyllody. The cause of their origin probably lies in hybridisation. Their study gives a general support to the close relationship of the family Papaveraceæ with the other families of the Rhoeadales, especially the Capparidacæ and to the hypothesis of pteridospermous origin of angiosperms.

Literature Cited.

1. BENSON, M. & WELSFORD, E. J.—The Morophology of the Ovule and Female Flower of *Juglans regia* and a few allied Genera. Ann. Bot. XXIII: 623–633, 1909.

2. COLE, R. D.—Imperfection of Pollen and Mutability in the genus Rosa. Bot. Gaz. 63: 110–123, 1917.

3. COULTER, J. M. and CHAMBERLAIN, C. J.—Morophology of Angiosperms. New York, 1903.

4. DEVRIES, H.—The Coefficient of mutation in *Oenothera biennis* L. Bot. Gaz. 59: 169–196, 1915.

5. DORSEY, M. J.—Pollen development in the grape with special reference to sterility. Minn. Agric. Exp. Sta. Bull. 144, 1914.

6. DUTHIE, J. F.—Flora of the Upper Gangetic Plain. Vol. I, part I, Calcutta, 1903.

7. DUTROCHET, H.—Sterility of Hybrid plants. Gard. Mag. 8: 500, 1832 (Quoted by Cole, 2).

8. ENGLER, A., and GILG, A.—Syllabus der Pflanzenfamilien. Berlin, 1924.

9. GAERTNER, C. F.—Versuche und Beobachtungen über die Bastarderzeugung in Pflanzenreich. Stuttgart, 1849.

10. GUERIN, P.—Le tegument seminal et les trachees nucellaires des Thymelaeacees. C.R. ac. Paris. 156: 396–400, 1913.

11. HOAR, C. S.—Sterility as the result of hybridisation and the condition of the pollen in Rubus. Bot. Gaz. 62: 370–388, 1916.

12. HUTCHINSON, J.—The Families of Flowering Plants. I. London, 1926.

13. JOSHI, A. C.—Megaspore-formation and Embryosac of *Argemone mexicana*, Linn. Jour. Ind. Bot. Soc. XII: 83–91, 1933.

14. JEFFREY, E. C.—Spore conditions in hybrids and the mutation hypothesis of DeVries. Bot. Gaz. 58: 322–336, 1914.

15. JUEL, H. O.—Beiträge zur Kenntnis der Tetraden-teilung. Die Tetradenteilung bei einer hybriden Pflanze. Jahrb. Wiss. Bot. 35: 638, 1900.

16. KERSHAW, E. M.—The Structure and Development of the Ovule of *Myrica Gale*. Ann. Bot. XXIII: 353–362, 1909.

17. ————Further observations on the structure of the Ovules of Myricaceæ and Allied Groups. Ann. Bot. XXIII: 692, 1909.

18. LOTSY, J. P.—Evolution by means of Hybridization. The Hague, 1916.

19. ROSENBERG, O.—Uber die Tetradenteilung eines Drosera-Bastardes. Ber. Deutsch. Bot. Gesells. 47: 53. 1904.

20. SEWARD, A. C.—Plant Life through the Ages. Cambridge, 1931.

21. THOMAS, H. H.—The Caytoniales, a new group of angiospermous plants from the Jurassic rocks of Yorkshire. Phil. Trans. Roy. Soc. Ser. B. 213: 299–363, 1925.

22. THOMAS, H. H.—The Early Evolution of the Angiosperms. Ann. Bot. XLV: 647–672, 1931.

23. TISCHLER, G.—Zellstudien an sterilen Bastard-pflanzen. Arch. Zellforsch. 1: 33–151, 1908.

24. WETTSTEIN, R.—Handbuch der Systematischen Botanik. Vienna, 1924.

25. WORSDELL, W. C.—The Principles of Plant Teratology. Vol. I. London, 1915.

Explanation of the Plate.

A photograph of two branches of *Argemone mexicana*, Linn. bearing abnormal flowers. About natural size.
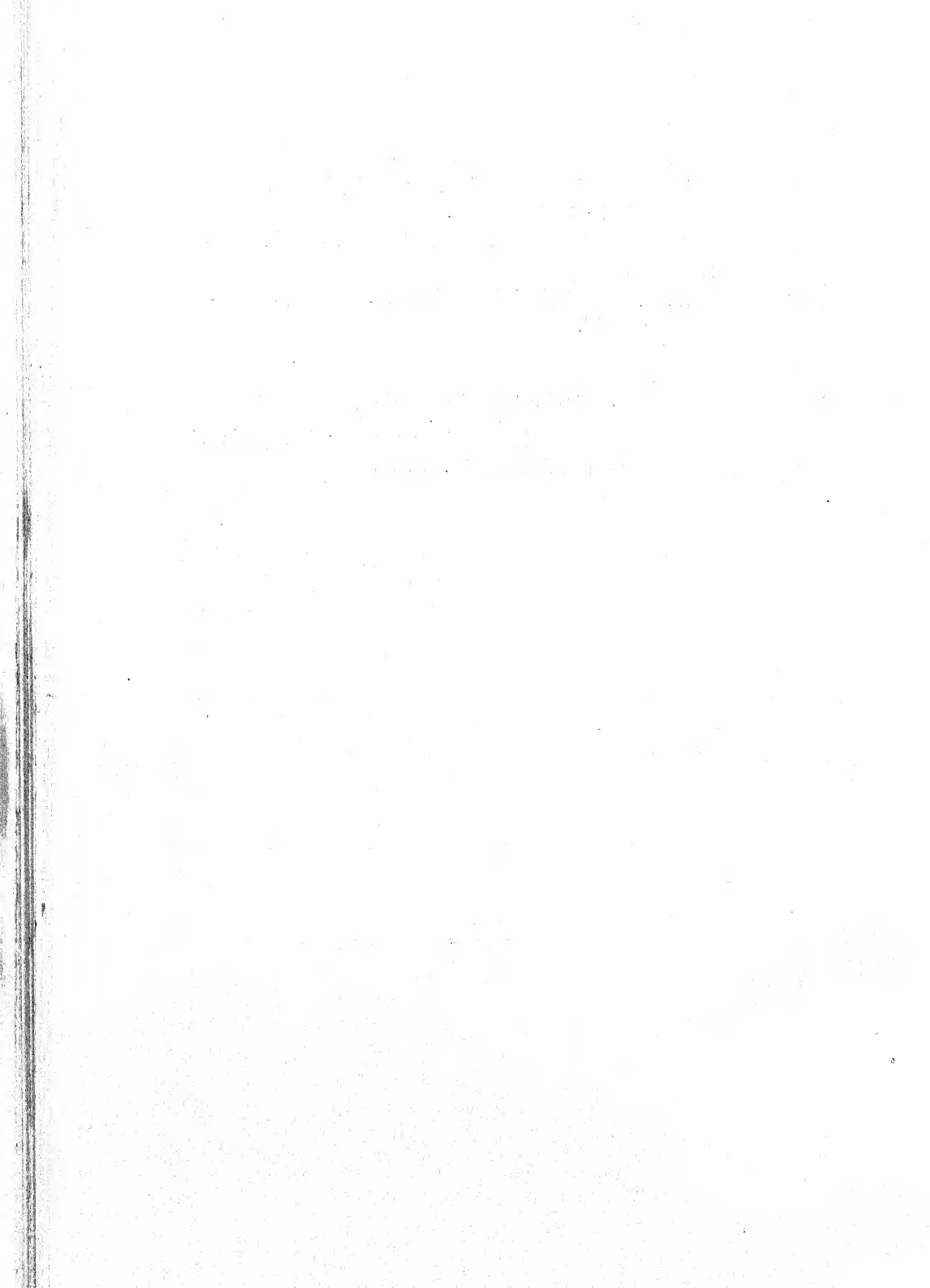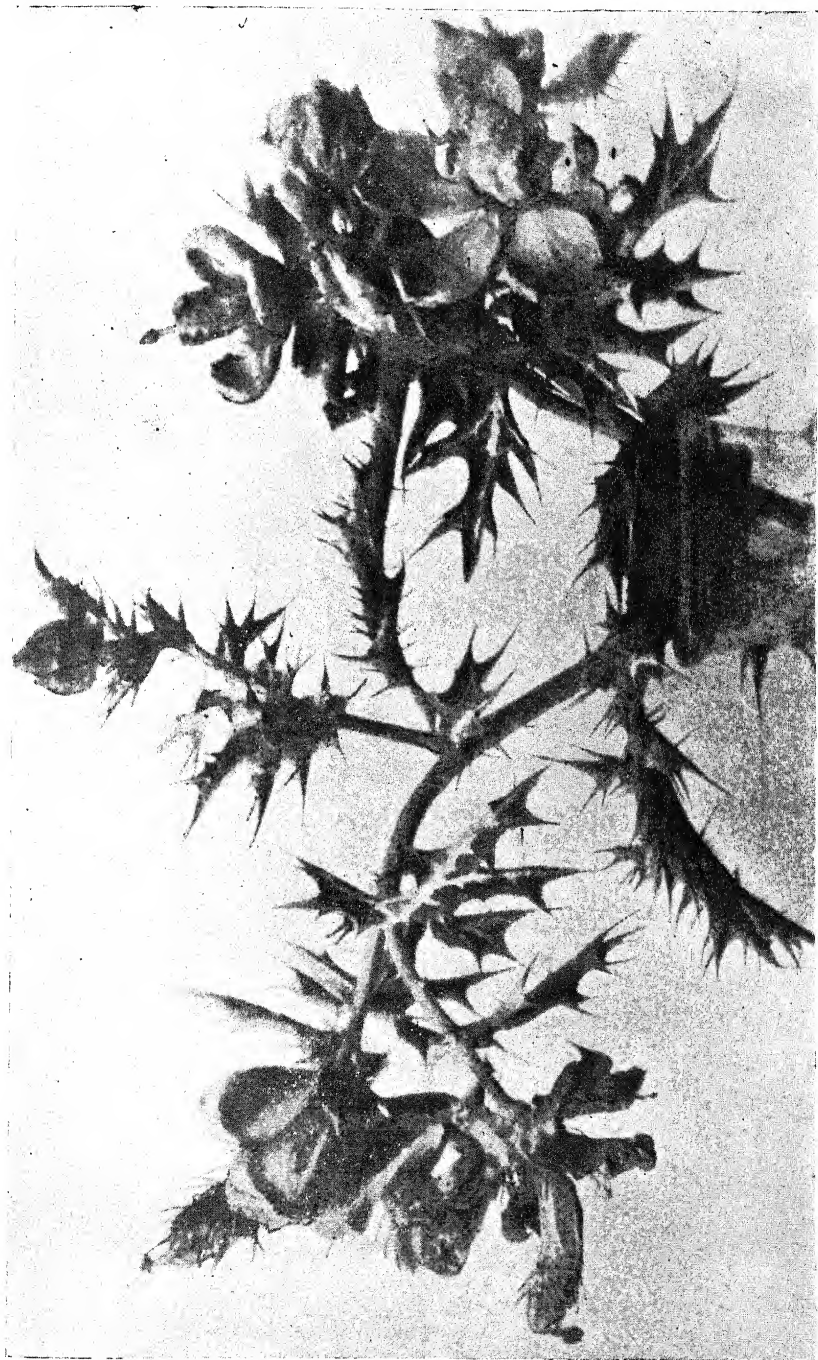
Fig. 10.

CHANGES IN PLANTS DURING LOW TEMPERATURES.

III. Some Chemical and Anatomical Changes in Plants grown under Controlled Temperatures.

BY

R. C. MALHOTRA,

Malhotra Street, Jammu, Kashmir State.

Freezing injury to fruit trees and other horticultural plants is sometimes so severe, at least in temperate climates as possessed by the northern portion of Kashmir State, that farmers suffer heavy losses (2). Occasionally even the sub-tropical areas suffer from this malady. For instance, orchards in the Bombay Presidency experienced rather an unusual degree of cold during 1929. As a result of this intensity of the falling of temperature, many fruit trees were killed or injured. Thus it is no wonder, that much interest has been shown in plant hardiness within recent years. For details parts I and II may be referred to which have already appeared in this journal.

Many research workers seem to attribute plant resistance against winter injury to various chemical substances. Rosa (18) and Hooker (8) think that hardening of plants is due to an increase in pentosans. Murneek (16) suggests that hemicelluloses may bring about hardiness. Shutt (19) showed that decrease in moisture has a definite relationship with hardening. Some workers such as D'Arsonval (5) think, that hardened plants have a lower freezing point. Harvey (7) shows the importance of the sap reaction. He suggests that there is a direct relationship between alkinity and hardiness or acidity and freezing injury.

Knowlton and Dorsey (9) and recently Crane (4) have presented data to show that peach buds borne near the terminals of the shoots are more advanced, consequently more matured and hardy, than those near the base. The more rapid growth of the buds on the terminal portions of the shoots is suggested to be due to their higher moisture, nitrogen and sugar content than that of the basal buds. Since no other study of the plant nitrogen has been made, as far as the writer is concerned, it may be of some interest to include nitrogen with reference to hardiness.

From the data of Maximow (14) it seems, that in some cases, salts exert more protective effects against freezing than even sugars. Furthermore, some workers seem to think, that more concentrated solutions, even of mineral salts, freeze at lower temperatures than less concentrated solutions of the same salts. Such is daily experimentation of a Physical Chemist. Thus ash is worth studying in this problem.

Some workers think that variation in the histological structures of the plant cells may be more important in hardiness than chemical composition. With the exception of one, no study has been brought to the writer's attention in this connection.

Inspite of the enormous work done on hardiness, particularly in the North America and Europe, no definite trend of the mechanism of the death of plants due to cold and freezing is obtained. It is true that several workers advocate, that the plant cells with higher sugar content may be able to withstand more severe temperatures. This has been questioned so much within the recent years, because of the results gained by the more sensitive instruments and precise methods, that for all practical purposes, the botanists are divided into three different schools. It is the writer's conviction that sugars alone may be or may not be so important in enabling a plant to escape winter injury as the sum total of carbohydrates with the exception of inert polysaccharides, as celluloses for instance. The latter chemical substances have been studied in respect to their physiological rôle of the so called hard and soft wood of plants. However, the thesis and data are so extensive both in materials, conclusions and results as well in space that they may just as well appear in the next part of the papers of this series.

Materials and Methods.

Tomato, lettuce, cabbage and Hungarian kale seeds were obtained from the same parents. Twenty plants of each crop were allowed to grow in the green house of the University of Chicago at about 50°F (comparatively cool) and about 75°F (comparatively warm). Soil, sunlight, oxygen and moisture conditions were alike. At the end of five months, half of the plants were transferred from warm to cold and *vice versa*. A month later, 5 plants of each crop and of each type, that is, the ones grown at 50°F, 75°F, transferred from cold to warm and warm to cold, were cut just above the ground line, their moisture content determined by drying them to constant weight at 65°C, ground and

pulverized until the powders obtained could pass a sieve of 60 meshes per inch. Such a degree of fineness was sufficient as suggested by Malhotra's data (10).

Pentosans were determined by a modification of Pervier and Gortners' method as developed by Malhotra (11). Hemicelluloses were estimated by the method of the official Agricultural chemists (17). Total nitrogen was found by the Arnold-Gunning modification of Kjeldahl (13) and was calculated as total proteins by multiplying the amount of nitrogen with 6·25, a factor accepted by Fuller (5B). Ash was obtained by burning the samples in an electric muffle (the data for only two typical anaylses have been shown here). The analysis of each sample was carried out in duplicate, sometimes in triplicate for close check.

Sap was extracted under 90 pounds pressure without freezing or thawing the plants, in order to prevent any change in the hydrogen-ion concentration of the tissues. It has been shown by many plant physiologists that during this process some apparent change of acidity and alkinity takes place within the sap. The sap so obtained was divided into two lots, one being used for determining the freezing point and the other for the estimation of pH. Hydrogen-ion determination was carried out by means of quinhydrone electrode after Clark (3). The colorimetric method was considered unsatisfactory because of the green colour of the sap present in these plants. Clearing the sap might have changed its pH value. The freezing point was determined by Beckman's apparatus (6). It is desirable to note that shortly after reaching the freezing point the temperature rises again. For the most accurate results any temperature other than the actual freezing point should always be avoided.

The remaining two plants were used for anatomical study. Pieces, one-half inch long, were cut nine inches above the ground line. Free hand, cross and longitudinal sections about 10 u were prepared, using a freezing microtome. The sections were stained according to Chamberlain (1).

Presentation of the Data and Discussion

A. *Moisture.*—The percentage of moisture is given in table I. From the data it seems, that on the whole, moisture in plants grown at 50° and 75°F was about the same. Cabbage grown at 50° showed a little more increase in water content than that grown at 75°; while lettuce No. 2 appeared to be the same but lettuce No. I grown at 75° contained 0·8 per cent more water.

A similar relationship was shown by tomatoes grown at these temperatures. On the other hand, Kale grown at 75° showed some increase over that of 50°.

Table I. The Effect of Temperature on the Moisture Content of Plants.

NAME AND NO. OF PLANT		PERCENTAGE OF MOISTURE			
		At 30°	At 75°	From cold to warm	From warm to cold
Cabbage	1.	88.60	87.90	88.48	88.14
	2.	88.40	87.31	88.10	87.60
Lettuce	1.	89.30	90.10	89.46	91.94
	2.	88.90	89.89	89.11	89.50
Tomato	1.	89.10	90.70	87.02	92.09
	2.	88.90	89.00	87.90	88.95
Kale	1.	88.30	89.02	87.76	89.18
	2.	88.40	88.95	87.89	89.29

Plants transferred from cold to warm did not behave alike. Some decreased (Tomato and Kale), while others (Lettuce) remained practically the same. Plants changed from warm to cold showed a similar irregularity, although, on the whole, they had a somewhat higher percentage of water than those grown under any other condition, except cabbage. It seems, that increase or decrease in the moisture content of plants grown under warm and cold conditions, other factors remaining the same, as shown by these crops, is so small that the difference may fall within the experimental error.

B. *Pentosans.*—The pentosan results are shown in table II. Plants grown at 50°F show a higher percentage of pentosan than those grown at 75°. When transferred from cold to warm, they showed a decrease in pentosan; while the plants changed from warm to cold showed an increase. However, the amount was less

than that of the plants which were allowed to grow at 50⁰. It
also seems that plants transferred from warm to cold, show more
pentosan percentage (except cabbage) than those from cold to
warm.

Table II. Effect of Temperature on Pentosan of Plants.

NAME AND NO. OF PLANT		PERCENTAGE OF PENTOSAN			
		At 50⁰	At 75⁰	From cold to warm	From warm to cold
Cabbage	1.	2.16	0.03	0.41	0.54
	2.	2.11	0.08	0.97	0.45
Lettuce	1.	2.11	0.13	1.10	1.85
	2.	2.00	0.19	1.20	1.41
Tomato	1.	1.64	0.57	0.71	0.93
	2.	0.98	0.39	0.73	1.00
Kale	1.	2.17	0.61	0.60	0.67
	2.	2.44	0.32	0.68	2.11

The total dry weight of the crop grown at 75⁰ was, on the aver-
age, 105 per cent more than that grown at 50⁰. Thus even the total
pentosans in the plants grown at 50⁰ were higher than those grown
at 75⁰. On the other hand, total pentosans, on the average, were
higher in plants transferred from cold to warm and lower in
plants changed from warm to cold, when considered on the basis
of their total weights. In many studies undertaken previously,
this fact does not seem to have been brought out. The plants trans-
ferred from warm to cold were just as hard as the ones grown at
50 degrees throughout. If the percentage, rather than the total
amount of pentosans, is responsible for hardiness in plants, cer-
tainly this data does not seem to show as much. For the fuller
review of various works in this connection Malhotra (12 B) may
be referred to.

C. *Hemicelluloses.*—The data on hemicelluloses are present-
ed in table III. It seems that in some crops grown at 50⁰ (Cab-
bage No. 1 and Lettuce Nos. 1 and 2) the percentage of hemicellu-
loses is higher, while in others (Tomato and Kale) it is lower.
On the other hand, all plants transferred from warm to cold seem
to contain more than any of the other lots. It is hard to account
for such an effective increase of hemicelluloses under the changes
brought out by these temperatures. The writer has already indi-
cated, that hemicelluloses are not only the cell wall materials but
should be looked upon as the active protoplasmic contents. As
such (12) it may be that some metabolic change in the reserve
substance due to the variable thermal transformation may have
taken place. Until we learn more about the nature of hemicellu-
loses than that recently explained by Miss O'Dwyer, a worker co-
operating with the British Cotton Association, we shall advance
but very little in this particular phase.

Table III. Effect of Temperature on Hemicelluloses of Plants.

NAME AND No. OF PLANT		PERCENTAGE OF HEMICELLULOSES			
		At 50⁰	At 75⁰	From cold to warm	From warm to cold
Cabbage	1.	5.94	6.23	7.45	22.1
	2.	6.32	6.67	6.81	18.4
Lettuce	1.	5.64	4.41	8.34	20.1
	2.	5.67	3.97	6.47	22.5
Tomato	1.	14.70	9.80	6.65	22.9
	2.	20.4	8.35	7.93	16.2
Kale	1.	12.8	6.79	9.25	20.70
	2.	17.7	7.93	7.55	19.91

D. *Total Proteins.*—It seems from table IV, that the plants
grown at 75⁰ show more total protein than those grown at 50⁰.
Plants transferred from cold to warm seem to exhibit more protein

than those grown continually at 50⁰ as well as those transferred from warm to cold. On the basis of percentages, plants grown at 75⁰, also those which were changed from cold to warm, the total proteins seem to be practically the same (except Kale No. 2), although on the whole, the former shows less percentage of proteins than the latter. Of course, the same relationship holds true for total nitrogen also, since it can be calculated by dividing the protein figures with 6·25, a factor of which mention has already been made.

Table IV. Effect of Temperature on the Total Proteins of Plants.

NAME AND NO. OF PLANTS		PERCENTAGE OF TOTAL PROTEINS			
		At 50°	At 75°	From cold to warm	From warm to cold
Cabbage	1.	13.1	18.3	19.3	13.6
	2.	14.7	17.9	18.6	16.2
Lettuce	1.	8.2	16.7	14.3	7.5
	2.	10.6	15.8	15.1	10.7
Tomato	1.	12.9	21.2	20.2	14.8
	2.	12.8	19.1	19.2	17.8
Kale	1.	12.7	19.8	20.7	13.9
	2.	13.5	16.7	18.2	15.2

The data seem to show a great variability of total proteins within the crops even when grown under the same conditions of temperature. It has been ascertained more than once from the Plant Nutrition Laboratory of the University of California, Berkeley, California, U.S.A., that the proteins vary even within a single plant. If this be the case, one should not look for the amount of protein or the total nitrogen as any index of a practical signifi-

cance. Unless one goes deeper into the composition of the plant proteins, such as amino nitrogen, non-amino nitrogen and perhaps still smaller fractions, it is improbable to segregate the rôle played by the proteins in the plant hardiness. Such an attempt has been made by the writer and a good deal of the work is still in progress. It will be reported in due course of time.

E. *Ash.*—The figures for ash presented in table V, seem to indicate (except Lettuce No. 2, Tomato No. 1 and 2 and Kale No. 2) that, plants grown at 50⁰ have a higher ash content than those grown at 75⁰. These differences, however, are not very great. On the other hand, plants transferred from cold to warm or *vice versa* have the higher percentage of ash content. It points out, however, that individual crops allow selective permeability to different salts as pointed out by Malhotra (12). Many Russian workers including Maximow, whose work has already been quoted above, in the main, agree with the conclusions already arrived at by the present writer.

Table V. Effect of Temperature on the Amount of Ash Content.

NAME AND NO. OF PLANTS		PERCENTAGE OF ASH CONTENT			
		At 50°	At 75°	From cold to warm	From warm to cold
Cabbage	1.	4.86	3.62	1.47	1.19
	2.	4.63	3.78	1.26	2.42
Lettuce	1.	4.97	4.53	3.02	3.60
	2.	4.58	4.51	2.45	1.81
Tomato	1.	5.03	5.30	5.40	4.22
	2.	5.99	5.67	3.49	3.04
Kale	1.	4.31	3.71	3.87	3.61
	2.	4.28	4.22	2.49	4.95

F. *Hydrogen-ion Concentration.*—The figures for the hydrogen-ion concentration of the sap have been shown in table VI. It seems that pH values obtained are very constant (about neutral point) for plants grown at 50⁰, 75⁰ or for those transferred from cold to warm and *vice versa*. It seems that pH varies but little, at least not significant enough to diverge more than pH 8 or less than pH 6. In some unpublished work, Malhotra found that the pH value of the sap within and outside of a plant varies quite materially. If this be true, in a general way, one should under-rate the importance of the hydrogen-ion study, now occupying such an importance in the physico-chemical topics of plant physiology.

Table VI. Effect of Temperature on the Hydrogen-ion Concentration of Sap.

NAME AND NO. OF PLANT		HYDROGEN-ION CONCENTRATION			
		At 50°	At 75°	From cold to warm	From warm to cold
Cabbage	1.	6.2	6.8	6.8	6.8
	2.	6.3	6.4	6.8	6.7
Lettuce	1.	6.2	7.2	7.4	7.1
	2.	7.2	6.9	7.2	7.1
Tomato	1.	7.1	7.1	7.2	6.9
	2.	6.9	7.0	7.2	6.9
Kale	1.	6.9	6.9	6.9	7.1
	2.	6.4	7.0	6.9	7.1

G. *Freezing Point of Saps.*—It would seem from table VII, that in all cases, sap extracted from the plants grown at 50⁰ showed a lower freezing point than that grown at 75⁰. Sap from plants (cold to warm) have varying values when compared with plants transferred from warm to cold. No uniform relationship seems to have been found in so far as these experiments are concerned.

Table VII. Effect of Temperature on the Freezing Point of Plant Sap.

NAME AND No. OF PLANTS		FREEZING POINT OF PLANT SAP GROWN			
		At 50°	At 75°	From cold to warm	From warm to cold
Cabbage	1.	5.1	4.7	4.9	4.9
	2.	5.0	4.8	4.9	4.8
Lettuce	1.	5.2	4.9	4.9	5.0
	2.	5.2	4.8	4.9	4.9
Tomato	1.	5.0	4.6	4.8	4.7
	2.	5.0	4.5	4.9	4.9
Kale	1.	5.4	4.5	5.1	5.3
	2.	5.4	4.9	5.2	5.2

Note:—Read minus in all cases such as—5.2.

H. *Anatomical Features.*—From the study of the cross-section it appears, that in the main, plants grown at 75⁰ develop more diameter than those grown at 50⁰. This difference is due, very largely, to many large parenchyma cells of the pith in favour of plants grown at 75⁰. By microscopic measurement of the diameters of about 500 cells of this nature, it seems that these cells in plants grown at 75⁰ are, on the average, 35 per cent larger than those grown at 50⁰. However the intercellular spaces of the cells from plants grown at 75⁰ are rather large; while the cells of plants grown at 50⁰, although somewhat smaller in size are undoubtedly compact. The compactness of the cells grown at the lower temperature may, in consequence, enable a plant to withstand a still lower temperature as suggested by Molisch (15).

It was also noted by the microscopic examination of these sections that the walls of all kinds of cells obtained from the plants grown at 50⁰ were somewhat thicker than those grown at a higher temperature. Shall we then assume that cells with thicker walls can withstand more withdrawal of water, which so often

takes place during freezing injury, than similar cells with thinner walls? An answer to this question cannot be given satisfactorily unless one tears apart these walls with a known pressure by means of micro-dissection (micro-manupilication needles) and similar observations on cells with induced ice injury and a known pressure exerted thereby. Unless this direction is explored, it would be fruitless to expect that this study in itself would be sufficient to lead us to the mechanism of winter hardiness. This must be confessed, however, that the combined data should bring us closer to the solution of this problem from a practical standpoint.

Charts No. 1 and 2 illustrate the distribution of various substances and properties in the crops used as mentioned in the above paragraphs.

Summary.

1. Cabbage, lettuce, Hungarian kale and tomato plants were grown at 50°F and 75°F. Half of the plants were transferred from cold to warm and *vice versa*. Their moisture content, pentosans, hemicelluloses, total proteins, ash, hydrogen-ion concentration and freezing points were quantitatively determined. Their anatomical study also was attempted.

2. No definite conclusions can be drawn as yet in relation to these materials with plant hardiness—freezing injury. The various difficulties presented by this problem are discussed in the paper. Further data are being collected as to fats, total carbohydrates, protein fractions, detailed cellular and colloidal changes, micro-dissection and micro-chemical tests, which when completed, may throw more light on this problem, possibly along with these data.

Literature Cited.

1. Chamberlain, C. J.:—Plant histology. Rev. Ed. 1925.

2. Chandler, W. H.:—Fruit growing. Houghton Mifflin and Co. 1925.

3. CLARK, W. M.:—The Determination of hydrogen-ions. 2nd Ed. Williams and Wilkins Co. Baltimore. 1923.

4. CRANE, H. L.—Physiological investigations on the resistance of peach buds to freezing temperatures. West Virginia Agr. Exp. Sta. Bul. 236. 1930.

5. D'ARSONVAL, M.:—La pression osmotique et son rôle de defense contra le foid dans la cellule vivante. Comp. Rend. Acad. Sci. Paris 133: 84–86. 1901.

5B. FULLER, H. C.:—The Chemistry and analysis of drugs and medicines. p. 889, John Wiley and Sons. 1920.

6. GETMAN, F. H.:—Theoretical Chemistry. 4th Ed. 251–54. John Wiley. 1928.

7. HARVEY, R. B.:—Hardening process in plants and developments from frost injury. Jour. Agr. Res. 15: 83–112. 1918.

8. HOOKER, H. D.:—Pentosan content in relation to winter hardiness. Proc. Amer. Soc. Hort. Sci. 17: 204–07. 1920.

9. KNOWLTON, H. E. and DORSEY M. J. A study of the hardiness of the fruit buds of the peach. W. Virginia Agri. Exp. Sta. Bul. 211. 1927.

10. MALHOTRA, R. C.:—Effect of degree of pulverization and weight of samples on quantitative analysis with particular reference to plant tissues. Jour. Ind. and Eng. Chem., Anal. Ed. 2: 398–401. 1930.

11. ———A modification of Pervier and Gortner's method for the estimation of pentosans in plant tissues. Journ. Ind. Chem. Soc. 8: 51–58. 1931.

12. ———Permeability of iodine in some economic plants. Protoplasma (Germany). 12: 1–22. 1931.

12B.———Changes in plants during low temperatures. Part I. Oils synthesis at various elevations of the Himalayas and its physiological influence on the protoplasm of Cedrus deodara. Jour. Ind. Bot. Soc. 10: 293–310. 1931.

12C.———Microchemical study of hemicellulose in some plant cells wtih special reference to its distribution in the protoplasm. Jour. Biochem. Japan. 12: 341–49. 1930.

13. MATHEWS, A. P.:—Physiological chemistry. 5th Ed. 994–96. 1929. William Wood and Co.

14. MAXIMOW, N. A.:—Experimentelle und kritische untersuchun-

gen ueber das gefrieren und erfrieren der pflanzen.
Jahrb. Wiss Bot. 53: 327–420. 1914.

15. MOLISCH, H.:—Untersuchungen ueber das erfrieren der pflanzen. 1897 Jena.

16. MURNEEK, A. E.:—Hemicellulose as a storage carbohydrate in woody plants with special reference to the apple. Plant Physiol. 4: 193–212. 1929.

17. U. S. D. A.:—Official and tentative methods of analysis of the Assoc. of Official Agricultural Chemists. Washington D. C., 1925.

18. ROSA, J. T. Jr.:—Investigations on the hardening process in vegetable plants. Missouri Agr. Exp. Sta. Res. Bul. 48: 1929.

19. SHUTT, F. F.:—On the relation of moisture content to hardiness in apple twigs. Proc. and Trans. Roy. Soc. Canada, 9: 149–153. 1903.
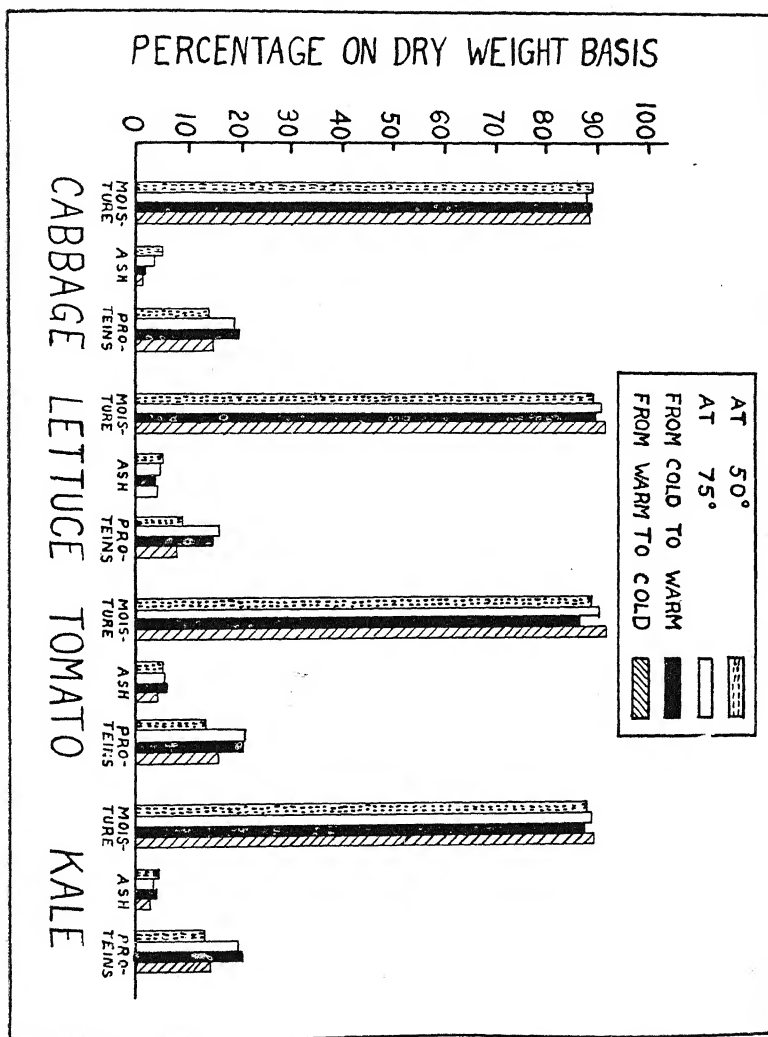
Chart No. 1 showing moisture, ash and the total protein contents in the various crops grown at different temperatures as well as those transferred from warm to cold and *vice versa.*
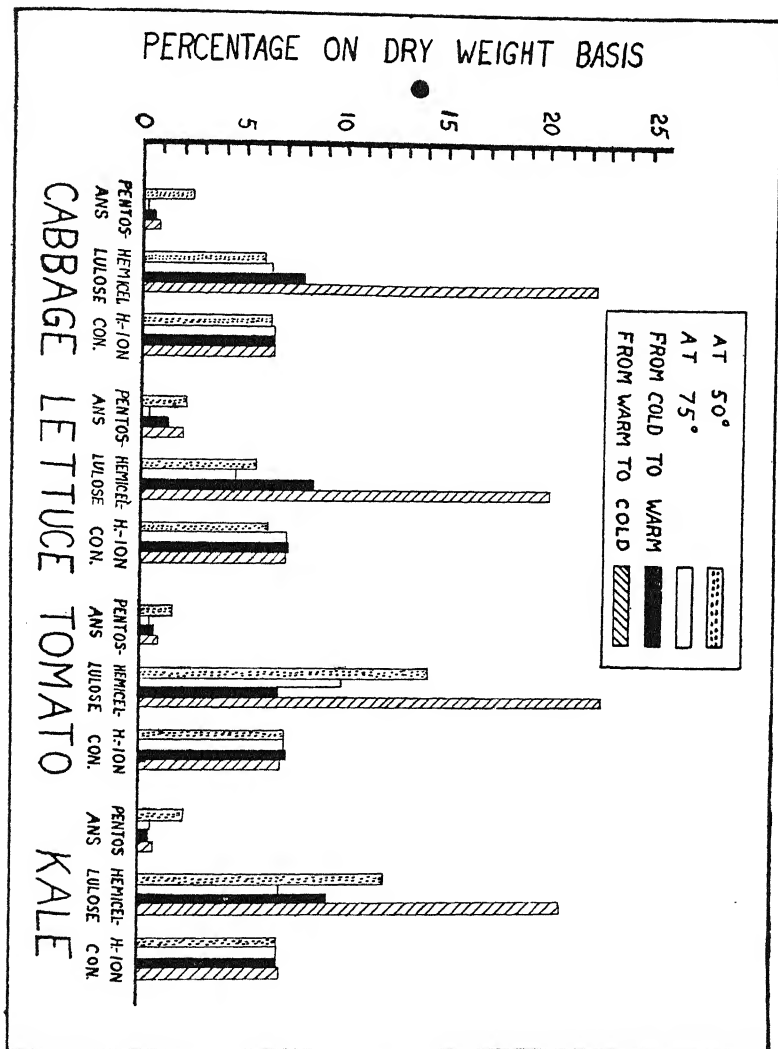
Chart No. 2 showing pentosans, hemicelluloses and the hydrogen-ion concentration in the various crops grown at different temperatures as well as those transferred from warm to cold and *vice versa.*

A NOTE ON THE EMISSION OF GLOBULES OF BASOPHILIC MATERIAL FROM THE NUCLEUS INTO THE CYTOPLASM DURING THE TELOPHASE OF NUCLEAR DIVISION.

BY

N. S. RAU, M.A.,

Udipi, South Kanara.

The view is now generally accepted that some kind of interchange of substance between the nucleus and the cytoplasm is constantly going on at all stages of cell-life (18), though not always in a manner which may be directly observed under the microscope. So long as the nuclear membrane is intact such an exchange can of course take place only by a process of diffusion through it, but during the final stages of the prophase of the nuclear division and in the following stages of metaphase, anaphase and early telophase no obstacles are found in the way of a free exchange of substance between the two parts of the cell, as the nuclear cavity is continuous with the cytosome during these stages. Not very much is known concerning the nature and the significance of such nucleo-plasmic exchange of substance on account of the difficulty of applying microchemical tests and of interpreting their results.

Under various circumstances, however, such as those of "physiological degeneration" caused by irregular conditions of nutrition (19) or in connection with certain phases of the nuclear division, the exchange of substance becomes more conspicuous and may be directly observed under the microscope in material prepared by the ordinary methods of histological technique. Nuclear substance thus cast out into the cytoplasm has been frequently described by many writers in a considerable variety of objects under the general name of "chromidia" or "chromidial substance". On the basis of such observations and by analogy with the conditions prevailing among certain potozoans, R. Hertwig (7) and others elaborated the "binuclearity hypothesis" as applied to the higher organisms, the chromidial substance being held to correspond to the "tropho-chromatin" supposed to be con-

7

tained in the macronucleus of the infusorians. At the present day this theory stands justly discredited, but the fact that nuclear substance may be extruded into the cytoplasm in a visible form as chromidia is well established; at one time there was some doubt with regard to the nature and the identity of these extruded bodies, but it has been established by the work of Duesberg (4), Schaxel (16), Hirschler (9) and others that they are nucleoproteid in their chemical composition and thus differ from aggregations of formed bodies found in the cytoplasm such as the mitochondria. Schaxel (15) has also put forward the view that such material of nuclear origin cast out into the cytoplasm may play an important part in the processes of cellular differentiation.

Such extrusion of chromidial substance in connection with karyokinesis, which is not by any means a matter of general occurrence, but has been recorded only in particular cases, has mostly been observed to occur in the early and middle phases of nuclear division. The writer has found, however, the emission of nuclear substance into the cytoplasm at a later stage in the course of nuclear division to be a matter of regular occurrence in all cases which he has had an opportunity of studying, both in the case of somatic mitosis and in that of the first and second meiotic divisions. During the middle and late telophase, when the chromosomes collect together at the poles of the cell and proceed to form the daughter nuclei, globules of strongly basophilic material stream out from the regions occupied by the chromosome groups into the cytoplasm (Figs. 1 and 2); the emission of these globules continues till a late stage of the telophase and they may be traced during the period when the dividing wall between the daughter cells completes its formation (Figs. 3 and 4). Finally they disappear in the general ground substance of the cytoplasm at a still later stage. The shape of these bodies is characteristically that of tiny globules, which suggests that the material is cast out as a fluid and not as a solid. They may be observed in material prepared with any of the fixatives that are commonly employed. such as the mixtures of Flemming, Bouin or Zenker. They stain deeply with the ordinary "nuclear" dyes such as safranine, gentian violet or iron-hæmatoxylin. Their affinity for these dyes, however, seems to be less strong than it is in the case of the chromosomes as the dyes may be completely extracted out of them with rather less difficulty than in the case of the latter; but this might well be due to the difference in physical conditions between the two, which in some measure determine, as A. Fischer (5), and

following him Bayliss (1), have pointed out, the precise character and intensity of any staining reaction. When these globules become dispersed in the cell and their substance gradually becomes mingled with the cytoplasm, it is found in suitably differentiated iron-hæmatoxylin preparations that the ground substance of the cytoplasm stains a darker grey than it does at other stages. "Acidic" stains such as orange or saure-fuchsin fail to stain these globules.

Judging from their place and manner of origin, it can be hardly open to doubt that the globules are derived from the chromosomes themselves: during the earliest stages of extrusion a crowd of them are always seen quite close to the chromosomes or even scattered among them (Figs. 1 and 2) and they become dispersed in the cell only at a later stage (Figs. 3 and 4). One may infer, therefore, that a stream of droplets of such basophilic substance issues forth from the chromosomes into the cytoplasm during the telophasic stages. It would, however, be an unwarranted conclusion to say that they are composed of "chromatin" merely because they give the same kind of staining reaction with some of the nuclear dyes as do the chromosomes; for, it must be remembered that none of these stains is specific for chromatin. What is extruded must be some substance which can have no place in the "resting" nucleus and is cast out accordingly when the chromosomes undergo the changes of structure and composition incidental to the formation of the resting daughter nuclei.

To the writer it seems probable that the extrusion of these globules of fluid material represents a phase in the cyclic exchange of nucleic acid compounds between the nucleus and the cytoplasm. Recent work makes it practically certain that the chromosomes lay the cytoplasm under contribution for part of the material required for their growth; this is specially evident in the case of cells undergoing the reductional division preparatory to the formation of reproductive cells. Reference may be made here to the papers by Latter (10) and by the present writer (14) where evidence pointing to the occurrence of such a transfer of material and to the part apparently played by the nucleolus in effecting it is set forth at some length. The fact that with the progress in the transfer of this material there is an increase in the intensity of the staining reaction of the chromosomes suggests that the substance thus added on must be the nucleic acid component rather than the permanent protein base of the chromatin. Careful bio-

chemical work, as for instance that of Masing (13) and that of van Herwerden (8) seems to show that the occurrence of the nucleoproteins is not confined to the nucleus, but that they, or at all events substances closely related to them and easily convertible into them occur in the cytoplasm as well and it is probable that during the nuclear division, these or their nucleic acid components are transferred to the nucleus to provide part of the material required for the growth of the chromosomes. This transfer, as pointed out already, seems to be effected through the agency of the nucleolus which plays an important part in connection with the storage and transfer of the chromatic substance throughout the history of the nucleus. For instance, in the course of nuclear division, during the "diffuse" stage in the growth period of auxocytes, when the chromosomes lose their basophily owing to the loss of nucleic acid, this substance becomes temporarily stored in the nucleolus, which takes a strong stain with basic dyes, and is restored to the chromosomes at a later stage. De Litardiere (11), working on ferns, has, by a study of the interrelationship between the chromosomes and the nucleolus, arrived at the conclusion that chromatic matter passes out of the chromosomes in the telophase and contributes to the formation of the nucleolus and is again transferred from the nucleolus to the chromosomes in the succeeding prophase, being mostly stored in the nucleolus during the metabolic stage of the nucleus, as was pointed out long ago by Flemming (6) and by Strasburger (17). A similar relation between the nucleolus and the chromosomes has also been found to exist by Martens (12), van Camp (2), Cleland (3) and other more recent writers who have studied the subject. It must be added, in the light of recent additions to our knowledge, that the cytoplasm is also probably involved in this cyclic exchange of nucleic acid compounds, contributing these to the nuclear components during the progress of the nuclear division. At the conclusion of the nuclear division the intranuclear nucleic acid becomes for the most part stored in the nucleolus, while that derived from the cytoplasm seems to be restored to it in the form of the globules described above. Fig. 4 illustrates the emission of such a substance in somatic telophase at a very late stage, only one of the daughter cells being shown. Figs. 1, 2 and 3 represent various stages in the first meiotic telophase of pollen mother cells in *Cyanotis cristata*, a plant in which, as in a great many others, the interkinesis is brief and the telophasic transformations of the nuclear components do not proceed very far; this plant has been

selected for illustration to show that even in such cases basophile material is cast out into the cytoplasm from the nuclear components.

The writer's object in publishing this preliminary note on the subject is to draw the attention of such of his fellow-workers as are in possession of facilities to conduct microchemical tests and pursue this line of investigation further, to a matter which has a bearing on the general question of the relationship between the nucleus and the cytoplasm, but which, probably on account of the somewhat inconspicuous character of the phenomena described, has escaped observation till now.

Literature Cited.

(1) BAYLISS, W. M., Lee's Microtomist's Vade Mecum, 1928, Chapter XI.

(2) VAN CAMP, G. M., La Cellule, 34: 7–48.

(3) CLELAND, R. E., Botanical Gazette, 77: 149–170.

(4) DUESBERG, J., Archiv für Zellforschung, 6: 40–139.

(5) FISHER, A., Fixierung, Färbung und Bau des Protoplasmas, 1899.

(6) FLEMMING, W., Zellsubstanz, Kern und Zelltilung, 1882.

(7) HERTWIG, R., Archiv für Protistenkunde, 1.

(8) VAN HERWERDEN, M.A., Anatomischer Anzeiger, 36: 193–207.

(9) ————Archiv für Zellforschung, 10: 431–449.

(10) HIRSCHLER, J., Archiv für Zellforschung, 9: 351–398.

(11) LATTER, J., Annals of Botany, 40: 277ff.

(12) DE LITARDIERE, R., La Cellule, 31: 255–473.

(13) MARTENS, P., La Cellule, 32: 333–428.

(14) MASING, E., Archiv für Entwicklungsmechanik, 31.

(15) RAU, N. S., Journ. Indian Botanical Society, 9: 79–113.

(16) SCHAXEL, J., Archiv für Zellforschung, 4: 265–308.

(17) ————Anatomischer Anzeiger, 39: 337–353.

(18) WILSON, E. B., The Cell in Development and Heredity, III Ed. p. 654.

(19) WILSON, E. B., The Cell in Development and Heredity, III Ed. p. 701.

Explanation of the Figures.

Fig. 1. Early telophase in the pollen mother-cells of *Cyanotis cristata*, oblique tangential section showing early stage in the emission of basophile globules. Flemming-iron-haematoxylin. × 1500.

Fig. 2. Later stage in the same species, oblique cross section, the globules are more scattered. Flemming-iron-haematoxylin. × 1500.

Fig. 3. Pollen mother-cell of the same species, later stage showing the formation of the transitory cell-wall following upon the close of the heterotypical division. The globules are few in number and reduced in size. Flemming-gentian violet. × 1500.

Fig. 4. Late telophasic stage in somatic mitosis in root-tip of *Vigna catiang*, only one of the daughter cells shown in polar view. Bouin-urea iron-haematoxylin. × 1500.
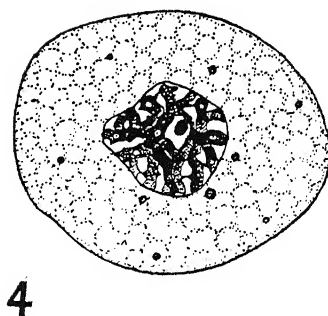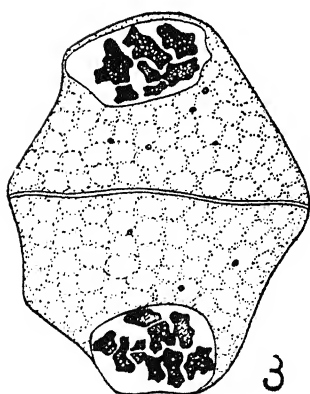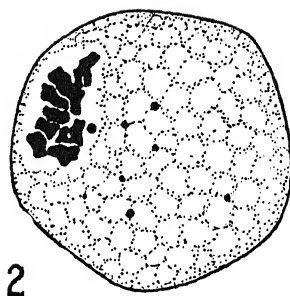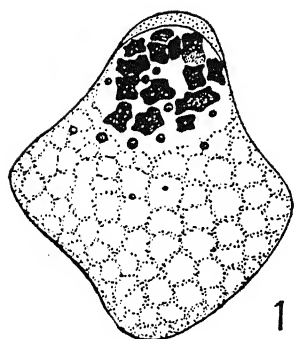
STUDIES IN ABSORPTION AND TRANSPIRATION
I. Cut Shoots treated with 20 per cent formalin

BY

T. Ekambaram and I. Madhusudana Rao.

Introduction.

It is true that physical theories are fast gaining ground and are accepted by many scientists for explaining the forces that are involved in raising water even to great heights in plants. Still the physiological theories for the ascent of sap have not been completely disproved. It is needless to trace out the progress of the two schools of thought from the beginning. Suffice it to say that at present the vitalistic theory stands strongly criticised by the Dixon School and that the latter's theory with little modification here and there finds the largest support.

According to Sir J. C. Bose, water is pumped up to the leaves by groups of pulsating cells situated in the cortical region along the stem. Dixon holds that the water in the vessels is always under high tension due to its cohesive property. Evaporation of water from the walls of the mesophyll cells in the leaves aided by the high suction pressure of these cells maintains a continuous current of water up the stem into the leaves. With all the data and experimental proof gathered in support of this theory, it has not yet been possible to prove definitely that the living cells of the plant have nothing to do with the ascent of sap. Thus it is that this portion of the work has been taken up to see whether the living cells (in the stem and in the leaves) play any part in the ascent of sap and if so, how they help in maintaining the transpiration stream.

Apparatus.

It was found that experiments conducted by other workers on this aspect were limited to the study of either transpiration or absorption. Rarely were they worked out together. Also the period of intervals for taking measurements was too long to notice some of the more important but constant changes occurring under experimental conditions. So it was felt that automatic records for the rate of absorption and for the rate of transpiration should be obtained; and the quantity absorbed each time or the amount of water evaporated should be as small as possible.

Apparatus was devised to satisfy the above needs. In the case of absorption, it has been possible to record ·05 c.c. of liquid absorbed each time; the quantity absorbed each time remains constant while the time varied. For transpiration, the time taken for evaporating 20mgms. of water is recorded.

I. Absorption

The apparatus (Fig. 1) consists of a U-tube, one limb of which is longer than the other. There is a side outlet tube (with a stop-cock
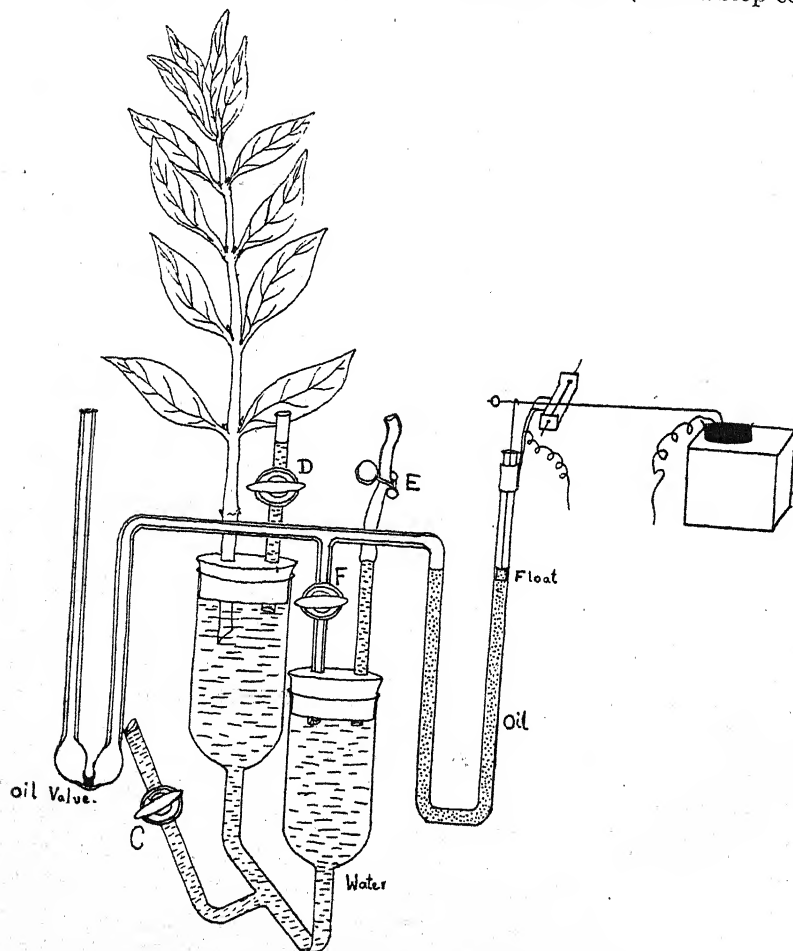
Fig. 1.

'c') starting from below. To the open end of the long limb of the U-tube is fixed a two-holed rubber stopper. Through one of these

holes is passed a glass tube with a stopcock 'd'. The open end of the short limb is also fitted with a two-holed rubber stopper. Through one hole is passed a glass tube open at both ends. Its upper end is closed with a piece of rubber tubing and a pinch-cock 'e'. Through the second hole is passed the lower end of the vertical tube of a T-shaped glass piece. It has a stop-cock 'f' in the vertical tube. The two ends of the horizontal part of the T-piece terminate respectively on one side into a 'bubbler' and on the other into a U-column. This glass piece excepting the U-column is made of narrow bore tubing.

There is a small drop of oil (Cocogem) in the bend of the 'bubbler' between the two bulbs. The U-column on the other side is filled up to three-fourths of its height with Cocogem oil. A small circular piece of cork (coated with an oil-proof varnish) is allowed to float in the oil (just below its surface). The float is attached to the short-arm of a balanced free-moving lever by means of a thin wire. The lever has a platinum point at the curved end of the long arm. The lever itself and its support are made of silver. Just below the tip of the long arm of the lever is a mercury cup resting on a wooden stand (not shown in the sketch) which can be raised or lowered. Two leads are taken off one from the mercury cup to one terminal of an accumulator, the other from the silver support of the lever to the electro-magnetic marking pen. The other terminal of the marking pen is connected to the remaining terminal of the accumulator. It is essential to keep a suitable condenser across the leads from the mercury cup and the lever to avoid slight sparking between the mercury surface and the platinum point of the lever.

Manipulation.

The stop-cock 'f' is closed. The stop-cocks 'c' and 'd' and the pinch-cock 'e' are kept open and the U-tube is filled with water. Now the stop-cocks 'c' and 'd' are closed. The cut end of the shoot is pushed gently through the hole in the rubber stopper fitted to the long limb of the U-tube. The cut end of the shoot is never exposed to air. While transferring it from the basin of water in which it is kept before fixing it up in the apparatus, the cut end is kept moist with a little cotton soaked in water. The junction of the stem with the upper surface of the stopper is sealed thoroughly with grafting-wax of low melting point. Now the pinch-cock 'e' is closed and the stop-cock 'f' is opened. There

should not be any leakage in the apparatus. During the experiment the apparatus was immersed in a water-bath up to the upper surface of the stopper fitted to the long limb of the U-tube, taking care that the open ends of the bubbler and the U-column are above the water surface.

The stop-cock 'f', when it is opened, keeps the bubbler and the U-column in communication with the U-tube. As the shoot absorbs water through the cut end, pressure inside the U-tube is reduced and the oil drop in the bend of the bubbler tends to move towards the 'inside' bulb. Simultaneously with this movement is seen the lowering of the oil-level at the open end of the U-column. The cork-piece floating in the oil also moves downward with the oil-level, pulling the short arm of the lever down. The platinum point at the other end of the lever moves away from the mercury surface. When the pressure inside the U-tube reaches a certain minimum depending upon the resistance offered by the oil drop in the bubbler, a bubble of air breaks through the oil valve allowing a certain amount of air into the system to equalise the pressures inside and outside. With this equalisation of pressures, the oil level in the open end of the U-column rises back to its original position carrying with it the float. The long arm of the lever comes down and its tip touches the mercury surface below. This completes the circuit and the electro-magnetic pen marks a dot on the graph paper wound round a drum moving at a known uniform speed.

The oil valve, after breaking, flows down and closes up the bend again. Once again due to absorption of water by the shoot, the oil goes up; the short arm of the lever is pulled down thus breaking the circuit and the process of making the contact with the breaking of the oil drop in the bubbler is continued. It is thus evident that the quantity of water absorbed each time is constant depending upon the quantity and nature of the oil used in the bubbler.

A number of determinations were made of the volume of water absorbed each time by means of a capillary tube (calibrated to ·001 c.c.) attached to the open end of the bubbler with a piece of rubber tube and kept in a perfectly horizontal position. A small drop of oil is sent into the capillary tube from the open end. When the bubbler is working, the movement of the oil-index in the calibrated tube for each dot on the recording drum is noted down. A mean of 5 readings is taken every time and this is

verified on different occasions. This calibrated tube is removed after obtaining the value of the bubbler.

A general idea of the apparatus and the mode of working have been given. One advantage about the apparatus is that the liquid inside the U-tube can be changed very quickly (within 5 minutes) with the help of the outlet-tube below, without disturbing the apparatus in any other way. The outlet tube is provided with a long rubber tube and is filled with the same liquid as the U-tube is filled in the beginning of the experiment. Now, for changing the liquid in the U-tube, the stop-cock 'f' is closed. After opening the stop-cock 'd' and the pinch-cock 'e', the liquid inside the U-tube is siphoned out through the outlet tube by opening the stop-cock 'c'. The U-tube is completely refilled with the necessary liquid through the same outlet tube and the stop-cocks 'c', 'd' and the pinch-cock 'e' are closed. The stop-cock 'f' is turned open and the recording is continued.

2. Transpiration.

The principle involved in this apparatus (Fig. 2) is not a new one. The transpiring shoot with its cut end in water is kept in one pan of a delicate balance. Weights are added to the other pan to bring the pans into equilibrium. Due to transpiration from the shoot, the pan carrying the shoot will be continuously losing weight. Each time this pan loses weight equal to the weight of a steel ball, one ball is dropped into that pan thus bringing the two pans into equilibrium. Simultaneously a dot is marked by an electro-magnetic pen on a drum which is rotating at a known uniform speed. Steel balls of uniform size and weight are used in all the experiments. A metal piece is hung to the hook of the pan with the weights and is allowed to move in oil so as to damp the oscillations. Breeze is avoided as far as possible so that the balance is not disturbed.

For changing the liquids in the U-tube in which the shoot is fixed, or for any other treatment of the shoot, recording is stopped; the balance is brought to rest and after changing the liquid in the tube, both the pans are again equalised and recording is started. All this treatment takes about 5 to 10 minutes and this interval can be seen in the record from the length of the gap.

All data are presented as graphs along with tables of actual records. The number of dots representing the amount absorbed or amount transpired each time (which is constant throughout each experiment) are marked along the X-axis while the rate is marked on the Y-axis. Y-axis is graduated from top downwards, so that a

298 THE JOURNAL OF THE INDIAN BOTANICAL SOCIETY.

fall in the rate can be seen at once as a slope in the curve and *vice versa*. The actual time is marked along the curve. Any treatment
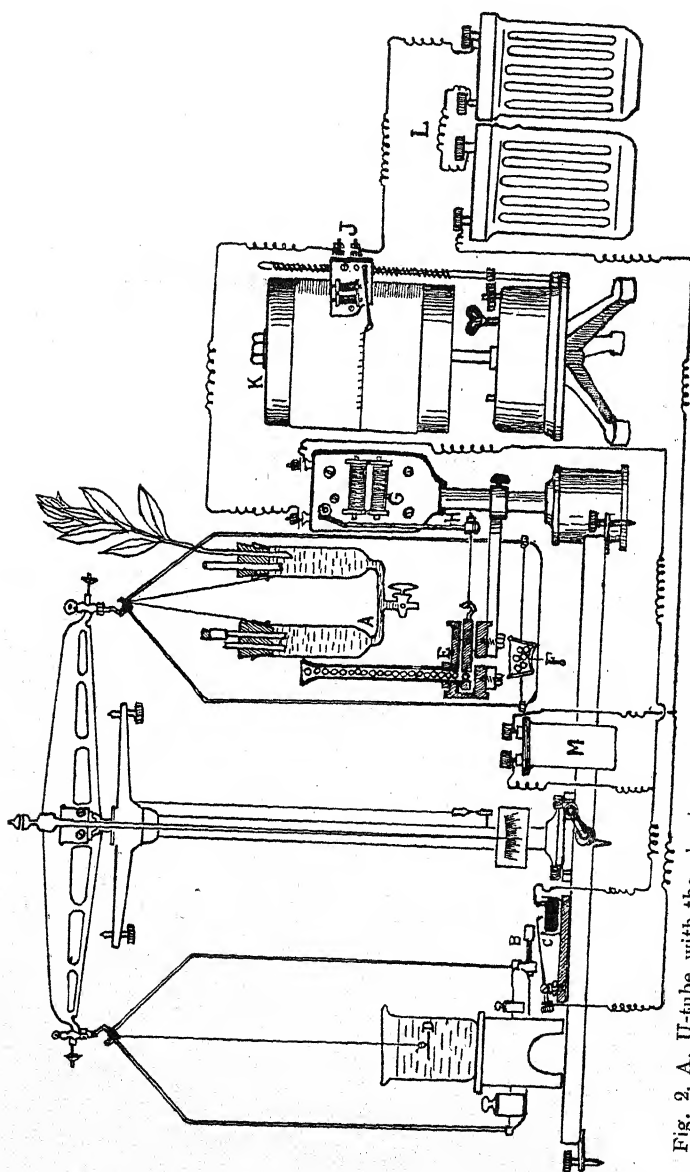
Fig. 2. A. U-tube, with the shoot mounted in one of the limbs with its cut end in water. B. A small wooden piece projecting from beneath the pan. C. Steel wire hanging beneath the wooden piece 'B'; the platinum point of the steel wire is just above the mercury in the cup below. D. 'Damper', moving in oil. E. 'Ball-dropping' apparatus. F. Receiver for the balls in the pan. G. Electro-magnet working the ball-dropper. H. Armature of the electro-magnet 'G', attached to the middle plate of the ball-dropper. J. Electro-magnetic marking pen. K. Rotating drum. L. Storage cells. M. Condenser.

to which the shoot is subjected is shown by an arrow-mark on the graph at that point and the actual time at that point is noted there.

Methods adopted.

After a preliminary study of the subject, the method resolved into a question of killing the cortical cells in the first instance and subsequent killing of the leaf-cells and studying the effect on absorption and transpiration. 20 per cent. formalin was used in all the experiments as this percentage was considered strong enough for killing the cells when even a small quantity was taken in.

Material used.

Initial difficulty was met with in the form of a big immediate fall in absorption when water at the cut end was replaced by 20 per cent. formalin. This was subsequently found to depend on the age of the shoot. Barleria cristata was chosen for study as shoots of the desired age were available for a greater part of the year. This plant is a perennial shrub, which, though in the summer months has a tendency to dry up almost, with the rains grows up vigorously putting forth new green shoots.

Shoots were cut carefully daily in the mornings at about 8 a.m. They were brought into the room and fresh cuts were made under water. The shoot was fixed in the apparatus and recording was started only after about an hour so that the shoot could in the meantime get over the effects of cutting and fixing in the apparatus. At the end of this period, the rate of absorption becomes constant. Recording was continued on almost all occasions throughout the day and night till next morning.

EXPERIMENTAL RESULTS.

Absorption.

The rate of absorption of water by the cut shoots was followed throughout the day and night on different occasions. The graphs obtained from these data show very little difference from one another in their general nature. So a control experiment was found to be quite unnecessary. One such experiment for the rate of absorption of water by a cut shoot is given in detail below.

Experiment No. 1. 25—4—31. Table 1; graph 1.

The shoot was cut at 7-30 a.m. and fixed up in the apparatus. Recording was started at 8-30 a.m. The rate was quite slow at the beginning but by 10 a.m., it was double the initial rate. Then till 3 p.m., there were slight variations. From 3 p.m., the rate was going down rather rapidly till 5 p.m. There was a slight pause till 6 p.m., when again the rate was decreasing continuously

till midnight. From 2 a.m., it was going up slowly though from 5 a.m., it was going up rather quickly. The experiment was continued till 6-45 a.m.

Description of the shoot:—$1\frac{1}{2}'$ in height; young; well-growing; 7 pairs of leaves; no branches. Temperature of the room;—30·9 degrees C., at 9 a.m. and 32·2 degrees C., at 4 p.m.

Experiments were conducted for rate of absorption under different treatments:

1. Water at the cut end was replaced by 20 per cent. formalin.
2. Leaves were coated on both sides with pure formalin.
3. The leaves were first coated with pure formalin and after about 3 hours, water at the cut end was replaced by 20 per cent. formalin.
4. Leaves were first coated with water. After the leaves were all dry, they were coated with pure formalin.

Typical experiments relating to these treatments are described in detail below:—

Experiment No. 2. 4—10—31. 20 per cent. formalin supplied to the cut end. Table 2; graph 2.

The shoot was cut at 8-30 a.m. and recording started at 9-30 a.m. At the beginning the rate was $6\frac{1}{2}$ minutes for absorbing ·05 c.c. of water; at 10 a.m., it was about $5\frac{1}{2}$ minutes. Just before adding the poison it was $4\frac{1}{2}$ minutes.

At 10-35 a.m., the poison was supplied to the cut end. The next dot was marked after 8 minutes. It took about 4 minutes for removing the water and adding the poison. At 11-30 a.m., when the rate was 13 minutes, the poison was observed in the top leaves by the slight discoloration along the veins. In another hour the discoloration of the leaves along the veins was complete. The rate of absorption at this time was at its minimum, *i.e.*, 27 minutes for absorbing ·05 c.c. At 1-15 p.m., the rate was recovering. It was about 22 minutes. At 5 p.m., it was 16 minutes. After this the rate was going down very slowly but never reached the rate of 27 minutes though at midnight it was about 23 minutes. The experiment was continued till 7-30 a.m., next morning. The leaves were all completely discoloured and were curling up by that time.

SHOOT:—10″ high; 6 pairs of mature leaves.

WEATHER:—Cloudy; cool.

TEMPERATURE at 11 a.m. Dry bulb—28·2 degrees C. Wet bulb—26·2 degrees C.

When 20 per cent formalin is supplied to the cut end, absorption keeps on at the same initial rate for about half an hour. Then there is a slow fall in the rate at which stage a slight discoloration of the veins could be seen in some of the leaves. The rate now goes down pretty quickly and the discoloration spreads to the veins of all the leaves. The rate reaches a minimum after about an hour and a half from the time of supplying the cut end with the poison. As absorption reaches the minimum rate, all the leaves have their veins discoloured. The rate then recovers but never reaches the initial rate. This recovered rate does not keep constant for a long time. It goes down slowly as the leaves turn brown completely and curl up.

Experiment No. 3. 6—4—31. Coating leaves with pure formalin.
Table 3; graph 3.

The shoot was cut at 7 a.m., and mounted in the apparatus in water. Recording was started at 9-40 a.m. The rate of absorption was almost constant except for a slight rise at 11-40 a.m.

11-40 to 11-55 a.m.—Coated all leaves with pure formalin.

The fall in the rate started gradually and reached its maximum at 12-20 p.m. Then the rate began to recover though not to the initial rate. The rate of absorption continued to be almost constant till 4 p.m., when it was again going down slowly.

SHOOT:—1½′ long; well-growing; 10 pairs of leaves.

TEMPERATURE:—32·5 degrees C. at 12 noon.

WEATHER:—Bright; warm; good breeze from 2 p.m.

When all the leaves of a cut shoot, whose cut end is kept in water, are coated with pure formalin on both sides, the rate of absorption undergoes changes. There is, in all cases, a fall in the rate; the fall begins usually from 15 minutes to half-an-hour. Generally the excess of formalin on the leaves is evaporated in about 15 minutes. The leaves rapidly turn brown and simultaneously, the rate of absorption goes down rapidly. It reaches a minimum in usually another half-an-hour. By this time all the leaves are completely discoloured. The curve here is going down rather steeply showing the rapid fall in the rate.

After this maximum decrease in the rate, it equally rapidly goes up again. This rise in the rate does not always finally equal the initial rate. It may be higher or lower but keeps fairly constant in almost all cases for some hours. Then it goes down very slowly so that the next morning the rate is quite low.

Experiment No. 4. 22—4—31. Coating leaves with pure formalin and later on supplying 20 per cent. formalin to the cut end. Table 4; graph 4.

The shoot was cut at 7-30 a.m. and mounted in the apparatus with the cut end in water. Recording was started at 9-20 a.m. The rate of absorption was a little irregular.

12 noon to 12-15 p.m.—Coated all leaves with pure formalin. 3 p.m.—Supplied 20 per cent. formalin to the cut end.

The rate of absorption was going down slowly for 15 minutes after coating the leaves with pure formalin. Later on the fall was much higher and the rate was at its minimum at about 1-15 p.m. The leaves were all discoloured. The recovery in the rate was equally rapid though the recovered constant rate was not equal to the initial rate of absorption.

When 20 per cent. formalin was supplied to the cut end, there was no fall in the rate.

The experiment was continued till 5-40 a.m.

SHOOT :—1½′ long; 9 pairs of young leaves; the shoot was vigorously growing; 4 small side branches.

WEATHER :—Bright; warm.

TEMPERATURE :—33 degrees C. at 3 p.m.

By coating leaves with pure formalin, the living cells in the leaves are put out of action. Now if 20% formalin is supplied to the cut end, according to Sir J. C. Bose, the 'pumping cells' in the cortical region are put out of action and there must be a fall in the rate of absorption. This fall in the absorption must be even greater than the one due to killing of the leaf-cells. There is no trace of such a fall. The record after supplying the cut end with 20% formalin follows the same course as in the case where no poison was supplied to the cut end.

Experiment No. 5. 5—3—31. Coating a long bare portion of the stem with pure formalin. Table 5; graph 5.

The shoot was cut at 8 a.m. and fixed in the apparatus, with the cut end in water. Recording was started at 10-40 a.m. The rate was rather slow but quite constant.

12 noon :—Coated 6″ of the bare stem, below the first node with leaves, with pure formalin. There was no fall in the rate of absorption till 4 p.m. The portion of the stem that was coated with formalin turned dark within an hour of coating.

Shoot :—1¼' long; mature; stem yellowish green; removed the leaves up to a height of 8"; about 2 dozens of small leaves with a few small branches at the top.

Weather :—Cool; cloudy.

In this experiment, the part of the stem turns brown within an hour of coating with pure formalin, indicating that the cortical cells are killed but the rate of absorption is not affected.

Experiment No. 6. 13—3—31. Coating leaves with water first and
        later on with pure formalin. Table 6; graph 6.

The shoot was cut at 8 a.m. and mounted in the apparatus with the cut end in water at 9 a.m. The rate of absorption was rather irregular.

12-20 p.m. to 12-30 p.m.—Coated all leaves with water. There was an immediate fall in the rate of absorption, reaching its maximum in about half an hour by which time all the water had evaporated from the surface of the leaves. The rate immediately recovered to initial rate and kept more or less constant.

2-35 p.m. to 2-50 p.m.—Coated all leaves with pure formalin. The experiment was continued only till 4-45 p.m.

When the leaves were coated with pure formalin, the rate was fairly steady for about half-an-hour. Then it began to go down. The rate was at its minimum at 3-45 p.m. The rate recovered to some extent in the next half an hour. It will be noted that the fall brought about by treatment with formalin did not reach the same magnitude as the one due to water-coating.

Shoot :—1½' long; first 4 internodes long; 4 pairs of big leaves; 5 pairs of tender ones.

Weather :—Bright; warm; no breeze.

When the leaves are coated with water, transpiration is almost completely stopped. This brings about a rapid fall in the rate of absorption. But as the water on the leaves is evaporated, normal conditions are restored. The rate of absorption, as the moisture on the leaves is evaporating, comes back to the initial rate equally rapidly. The curve is very steep on both sides. By the time the leaves become dry, the rate of absorption reaches the initial rate and follows the original course.

When the leaves are coated with pure formalin, there is no such immediate reduction in absorption. The rate of absorption continues to be almost the same for about 10 minutes at least. The formalin from the surface of the leaves evaporates very rapidly and the surfaces of the leaves are dry within 15 minutes

8

of coating the leaves with formalin. During this interval, the rate of absorption is not much changed. With the drying of the leaf-surfaces discoloration is seen here and there. A visible fall in the rate starts with this discoloration of the leaves and the rate reaches a minimum in another half-an-hour when the leaves are completely discolored. Now the rate of absorption begins to recover, sometimes quite rapidly. The recovered rate is generally lower than the initial rate.

Transpiration.

Experiments on the rate of absorption under various treatments have been considered till now. Similar experiments were conducted on the rate of transpiration also by shoots of the same plant.

The shoot was cut in the morning at about 8 a.m. and the cut end was kept immersed in water in the apparatus. Rate of transpiration from the shoot with its cut end in water was observed on different days and the graphs obtained from these data are quite similar to one another in their general nature. One such experiment is described in detail.

Experiment No. 7. 5—2—31. Rate of transpiration with the cut end in water. Table 7; graph 7.

The shoot was cut at 7-15 a.m. and mounted in the apparatus with the cut end in water. Recording was started at 8 a.m. The rate of transpiration was almost constant except for slight variations till 3 p.m. when there was a slow fall. At 4 p.m. the rate was one-third the rate at 8 a.m.

SHOOT :—1½' long; 6 pairs of leaves.

WEATHER :—Warm; bright.

TEMPERATURE :—26·8° C at 2-30 p.m.

Experiment No. 8. 7—1—31. Supplying 20% formalin to the cut end. Table 8; graph 8.

The shoot was cut at 8-30 a.m. and set up in the apparatus at 10 a.m. Recording was started at 11-55 a.m. The rate of transpiration was quite constant. At 12-15 p.m., 20% formalin was supplied to the cut end. From 12-30 p.m., there was a slow decrease in the rate of transpiration. At about 1-5 p.m., the rate was at its minimum. The rate was again becoming high. At 1-50 p.m., it was equal to the initial rate. The rate continued to quicken though rather slowly till the end of the experiment. By 3-30 p.m., all the leaves were completely discoloured.

Shoot:—The shoot was about 10″ high; a dozen leaves of medium size.

Weather:—Bright; clear.

The following points of difference between the effects of 20% formalin, supplied to the cut end of the shoot, on absorption and transpiration are clear from the above experiment:

1. The fall in the rate of transpiration starts within 15 minutes of supplying the poison to the cut end. There is no discoloration in any of the leaves by that time.

The fall in the rate of absorption starts only after about half an hour; signs of discoloration are seen in some of the leaves.

2. In transpiration, the minimum rate is obtained always within an hour of supplying the poison. The discoloration is not clear in all the leaves.

In the case of absorption, the minimum rate is obtained only after about 2 hours, when all the leaves are completely discoloured.

3. The minimum rate in transpiration is about half the initial rate while in absorption the minimum rate is less than one-third of the initial rate.

4. The recovery in the case of transpiration is complete at a time when the absorption shows a minimum.

5. The recovered rate in transpiration is either equal to or even higher than the initial rate; while in absorption, it is less than the initial rate.

Experiment No. 9. 17—1—31. Coating leaves with pure formalin. Table 9; graph 9.

The shoot was cut at 8 a.m. and fixed in the apparatus with the cut end in water. Recording was started at 9 a.m. The rate was not quite constant. It was going down slowly.

At 10-45 a.m. recording was stopped and the leaves were coated with pure formalin. It took about 15 minutes to start recording again. When the leaves were all coated with pure formalin, there was excess of formalin which had not evaporated before the recording was resumed. So the rate of loss of weight in the beginning was higher due to the evaporation of this excess formalin on the leaves. It took about 15 minutes for the leaves to get dry of formalin. Leaves were all completely discoloured by 12 noon and the leaves were curling at 1 p.m. By the end of the experiment, the leaves were all shrivelling up.

The discoloration of the leaves starts within 15 minutes after coating the leaves and all the leaves are discoloured in less than an hour. During all this time the rate of transpiration is equal to the initial rate or even higher. But with the curling of the dead leaves, a gradual fall in the rate is noticed and this occurs about four hours after coating the leaves with pure formalin.

In the case of absorption, the fall in the rate starts with the beginning of discoloration in leaves and reaches a maximum with the complete discoloration; then the rate recovers to a certain extent and this recovered rate keeps constant for some hours. When the dead leaves are shrivelling up, there is a gradual fall in the rate of absorption also.

The following experiment gives us an idea of how the rate of transpiration goes down when there is no supply of water from below.

Experiment No. 10. 30—1—31. Water-supply at the cut end withdrawn after some time. Table 10; graph 10.

The shoot was cut at 7-30 a.m. and fixed in the apparatus with the cut end in water. Recording was started at 8-30 p.m. In the first half-an-hour, the rate was irregular. At 10 a.m. water was withdrawn from the apparatus leaving the cut end exposed to air. There was a slow fall in the rate till 10-30 a.m. when it became more rapid. Visible drooping of leaves was clear even at 10-15 a.m.

SHOOT:—1′ long; 12 leaves. Bright weather.
TEMPERATURE:—27·3°C at 2-30 p.m.

There is much contrast between the appearance of a shoot which is killed by supplying 20% formalin at the cut end and that of another whose water-supply is withdrawn. In the first case, the leaves retain their original position even after the poison reaches them. The leaves get completely discoloured but still they do not lose their turgidity completely. They are not drooping. Finally they turn completely brown, and begin to curl.

In the case of the shoot which was deprived of its water-supply at the cut end, there are visible signs of the leaves drooping within the first 15 minutes of removing the water supply. The shoot tip slowly bends down; the leaves get more and more flaccid; they finally droop down completely and hang loose.

Behaviour of Stomata.

It was found essential to note the change in the stomatal apertures in leaves when poison was supplied to the cut end.

Experiment No. 11. 1—9—31. Behaviour of stomata when 20% formalin was supplied to the cut end. Table 11.

Porometer readings were taken of two shoots, one a control and the other, the experimental one. Porometers were attached to the leaves (lower side) by means of plasticine. The lower ends of the porometers were kept in water. Water was sucked up to a particular height in each case and the time was noted for a fall in the water-level through ·5 cm. After taking the readings for about 2 hours, water was replaced with 20% formalin at the cut end of the experimental shoot. The cut end of the control shoot was in water.

There is no marked change in the stomatal apertures of the leaf of the experimental shoot until it begins to get discoloured by the poison. When the leaf is discoloured, the stomata are comparatively open. Leaving aside the minor variations of the stomatal apertures, it can be noticed that the poison has a definite effect upon the stomatal openings. When the leaf-cells are all killed and thus the stomata also, the stomata are kept open. The open stomata allow an easy evaporation. Thus the leaf-cells when they are killed lose their fresh weight pretty quickly.

Recapitulation of Results.

Rate of absorption of water by a cut shoot of Barleria cristata (Expt. 1) is more or less constant from morning 10 a.m. till 3-30 p.m. when there is a definite fall. Till 3-30 p.m. the rate oscillates within small limits. The fall which begins at 3-30 p.m. becomes rapid gradually but at about 6 p.m., the rate gets a little steady for about half an hour. Again there is a rapid fall, the rate reaching a minimum at about midnight. From 2 a.m., the rate begins to recover and from about 5 a.m., the recovery is rapid.

Rate of transpiration (Expt. 7) from a cut shoot of Barleria with the cut end in water is more or less constant from 8 a.m. till about 3 p.m. when there is a gradual fall.

When the supply of water at the cut end of the shoot is withdrawn, the rate of transpiration (Expt. 10) from the shoot undergoes a definite change. For the first 15 minutes of the withdrawal of water-supply, the rate follows the same initial course,

but there is a continuous rapid fall afterwards. Visible drooping of the leaves is clear with the beginning of the fall in the rate of transpiration and at the end of 2 hours, all the leaves are drooping completely and the shoot tip also bends down.

Other experiments were conducted to note what changes are brought about in absorption and transpiration and the condition of the shoot, by killing various parts of the shoot. The cortical tissue throughout the stem was killed first and afterwards the leaf-cells by sending in a current of 20% formalin through the cut end of the shoot. The fall in the rate of absorption (Expt. 2) is seen only when the leaf-cells are killed. The killing of the cortical cells does not bring about any marked change in the rate of absorption. When all the leaves are completely killed, the rate reaches a minimum. After this stage, the rate of absorption recovers to a certain extent and this recovered rate keeps constant for some hours. In the case of transpiration with the same treatment of the shoot, the rate (Expt. 8) goes down as the poison reaches the leaves. With the killing of the leaf-cells, the rate recovers and this recovered rate is equal to or sometimes higher than the initial rate. The fall in the rate obtained in the case of transpiration as the poison reaches the leaves, is negligible compared to the fall in the rate of absorption obtained as the leaves are all killed by the poison.

In the next set of experiments, where the leaf cells alone are killed by coating the leaves on both sides with pure formalin, the rate of absorption (Expt. 3) begins to go down from 15 minutes after the treatment and reaches a minimum within an hour. The rate recovers immediately and this recovered rate keeps constant for at least 4 hours. In the case of transpiration (Expt. 9) with the same treatment, the rate is kept up at the same initial rate or even higher for about four hours. A slow fall in the rate is then noticed with the curling of the dead leaves. In these experiments the cortical cells of the stem are not affected.

In another set of experiments, the leaf-cells are first killed by coating them with pure formalin. The same changes in absorption as seen in the previous case are obtained. When the rate has completely recovered, the cortical cells are killed by sending a stream of 20% formalin through the cut end. The absorption (Expt. 4) is not affected in any way with the second treatment. The course of absorption followed that of the previous experiment (Expt. 3) where the cortical cells were not killed.

In the experiments considered till now, the cortical cells are killed by sending a stream of 20% formalin through the cut end. In Experiment No. 5 the cortical cells of a long portion of the stem are killed by coating that portion of the stem with pure formalin. The coated portion of the stem turned brown within an hour but there was no change in the rate of absorption even after four hours.

To understand the effect of coating leaves with pure formalin on absorption, the leaves of a cut shoot are first coated with water. Transpiration is almost stopped and there is an immediate fall in the rate of absorption (Expt. 6). When the water on the leaf surface had all evaporated, the rate equalled the initial rate. Now the leaves are coated with pure formalin. The fall in the rate of absorption does not follow immediately. With the evaporation of excess formalin on the leaf surface, the fall in the rate starts and with the complete discoloration of the leaves, the rate reaches a minimum. Then the rate recovers to a certain extent. That this fall in the rate of absorption is not mainly due to the water in the pure formalin is seen by the time taken for the fall to start. In all cases the fall starts after the evaporation of excess formalin on the leaf surface; but when the leaves are coated with water, the fall is immediate and the rate recovers by the time the surface of the leaf is dry.

The condition of the stomata (Expt. 11), when they are killed by sending in 20% formalin through the cut end, has also been studied by taking porometer readings. When the leaves are killed thus, the stomata keep fairly open. This fact has been confirmed by direct observation under the microscope.

Discussion.

It is interesting to note that the vitalistic theory started by Godlewski and followed up by Westermaier, Janse and many others is still supported by some scientists though with some modifications.

Ewart described some experiments to prove that the living cells of the stem are essential for ascent of water in trees. He repeated one of Strasburger's experiments, by cutting a tall tree at the base and keeping the cut end in formalin solution. He measured the rate of absorption in litres per hour. There was a fall in the rate till the second day. On the third day there was a rise in the rate and at 5 p.m. on that day the rate was half that noticed on

the first day. This rise in the rate and the continuous fall later on coincides with the results of our experiment where 20% formalin is supplied to the cut end. Ewart did not explain this rise in the rate of absorption amounting to 50% of the initial rate. He had to overlook this rise in the rate to say that a gradual fall was obtained in the rate of absorption as the living cells in the stem were killed when formalin was supplied to the cut end. The data obtained in this paper show clearly that this fall and subsequent rise in the rate of absorption are constant features and are due to changes brought about in the cells of the leaves by the poison and not to the killing of the cells in the stem.

Overton conducted a number of experiments with Cyperus to note the effect of killing long stretches of stem on the rate of transpiration and the condition of the leaves. He used steam, hot paraffin and poisonous fluids (e.g., picric acid, 95% alcohol, 40% formaldehyde) for killing the stem. He enclosed the portion of the stem to be killed in a glass tube and passed steam or poisons into it. With steam, he got a black poisonous substance exuded into the vessels which blocked them up partially. There was a gradual fall in the rate of transpiration. When this poisonous substance reached the leaf, there was discoloration of the leaf in patches, withering and final drying up of the leaf. He contends that though there is a fall in the rate of transpiration, there must not be a withering and drying of leaves. This withering and drying is due to the killing of the leaves and not to lack of water-supply. When he killed the stem with hot paraffin, the transpiration was not affected. Fresh leaves were also put forth after the treatment. So he says that steaming a portion of the stem causes an exudation of a poisonous gummy substance which blocks up the vessels partially and also when it reaches the leaves kills them.

When he used poisons, some of them never entered the vessels. In cases where the poison did not reach the vessels, the leaves continued to be fresh, and fresh branches were put forth.

In some experiments, he killed the whole shoot by keeping the cut end in a poisonous fluid (Mercuric chloride, Picric acid and Chromic acid) and when the poison was well seen in the leaves. he replaced it with water. The whole shoot was killed including the leaves. From the time of keeping the cut end of the killed shoot in water, he weighed the shoot with the water, once a day.

He had taken weights of a healthy control shoot also, with the cut end in water. He found that the killed shoot was losing more water than the control shoot. He continued weighing for 8 days. All these days the killed shoot was losing weight at a higher rate than the control. On the eighth day he noted the percentage of water in the killed shoot and also in the control shoot. In all cases he found that the percentage of water in the killed shoot was far below the percentage of water in the control shoot. He also noted that with different poisons, the rate of transpiration was different though in all cases it was much higher than the transpiration of the control shoot. In conclusion, he says that in these cases the tissues in the leaves are ruptured so as to expose additional cell-surfaces to the atmosphere. He further mentions that in the case of the poisoned leaves of Cyperus there can be no osmotic suction by the leaf cells. The imbibitional action of the cell-walls may, however, still keep the walls wetted and the suction caused by evaporation may be transmitted to the cohering water columns of the vessels. He concludes by saying "it is certain that in the case of a plant poisoned throughout, the elevation of the water in the stems and its evaporation from the leaves in larger quantities than normally occurs in living plants, depend purely upon physical forces".

It is interesting to note that he did not measure the rate of absorption but assumed that it might be equal to the rate of evaporation. The data obtained in this paper show that absorption is less than the rate of transpiration in a killed shoot. From the fact that the percentage of water in the killed shoot at the end of the experiment is less than that of the control shoot, Overton could have inferred that absorption was much behind evaporation. Results of the experiments described already show that there is a higher rate of evaporation from the killed leaves but simultaneously with this higher rate of evaporation a fall in the rate of absorption is noticed. This fall in the rate of absorption was not observed by Overton as he did not take measurements of absorption. It is also difficult to imagine how imbibitional forces of the cell-walls and evaporating power of the air can be transmitted through a mass of 'ruptured tissues' to the water in the vessels. The results of the experiments already described lead to the conclusion that the fall in the rate of absorption with the killing of the leaves and the subsequent rise in the rate are due to physical forces but different from those attributed by Overton.

MacDougal in his publication mentions that he has enough data to prove that the living cells of the stem have nothing to do with the ascent of sap. Only when the living cells of the leaves are killed, thus disturbing the osmotic strength of the sap in those cells, the pull from above on the water columns is affected and the absorption is consequently reduced.

Dixon says that, when a poison is supplied to the cut end, there is a sudden fall in the rate of transpiration when the poison is seen in the leaves. It is not so in the experiments described here. There is no such big fall though a slight fall was noticed before the leaves were discoloured. According to Dixon, when the poison reaches the leaf-cells and kills them, their osmotic properties are destroyed; they lose their turgor and there is a general collapse of the cells. But in the experiments described here, there is a clear difference in the effects of poison on the rate of absorption and rate of transpiration, especially in the first few hours. The fall in the rate of absorption is seen only when the leaf-cells are killed but when this happens, there is no fall in the rate of transpiration; either it is maintaining the initial rate or it is somewhat higher.

Sir J. C. Bose finds that pulsatory activity of cells in the cortex pumps the water up through the stem. When this activity is interfered with by any poison, the absorption is reduced and when all these pulsating cells are killed completely, there is no more ascent of sap. Most of his conclusions are based on observations of the changes in the leaf-position. But he gives two experiments for proving his assumption definitely. When 25% formalin was supplied to the cut end of a shoot, (shoots of Chrysanthemum and Mango were used by him) there was a gradual fall in the rate of absorption, the fall becoming considerable in the fourth and fifth hours after which there was no more absorption. He took readings once an hour. 20% formalin was used under similar conditions in the experiments described here. The fall, it was noted, starts in half an hour and reaches a maximum in two hours by which time the leaves are all discoloured; then the rate is observed to recover to a certain extent and the recovered rate keeps on for some hours. Till the leaf-cells are killed, there is no fall in the rate of absorption and absorption never falls to zero as stated by him. The fact that absorption is affected only by the killing of the leaves is proved by the Experiment No. 4.

Special attention is now drawn to one particular feature in the absorption and transpiration graphs. In these experiments, the fall in transpiration is noticed as the poison was reaching all the leaves i.e., much earlier than the fall in absorption which is obtained with the killing of all the leaf-cells. The fall in transpiration is much less in magnitude than the fall in absorption. Also at a time when the leaf-cells are all being killed, or when the rate of absorption is at a minimum, the rate of transpiration is equal or even higher than the initial rate. These facts have not been noticed by Dixon or other workers. From these data, it appears to be clear that at the time when the leaf-cells are killed, there is more water available for evaporation. This excess should have been released from the cells during killing. The effect of this is to keep transpiration at the same initial rate or even at a higher rate, though it reduces the pull on the water-columns in the vessels thus bringing about a fall in absorption. This change is only a temporary one and old conditions are being restored with the evaporation of the excess water in the leaf. There is a rise in the rate of absorption though it does not recover to the initial extent. This recovered rate keeps constant for some hours. A recovery in the rate of absorption after the leaf-cells are killed and its maintenance for some hours at the same level cannot be explained by Dixon's assumption that during the death of a cell, it collapses losing all its osmotic property. According to him, the tissues are all ruptured when they are killed. But the curves obtained from the experiments show that the mechanism for absorption or transpiration is not radically affected as postulated by other workers, when the leaf is killed with a poison. The dead leaf appears to retain its capacity for absorption and transpiration to an extent very near that of the living leaf.

Summary.

Apparatus for recording automatically the time taken for absorbing minute quantity of liquid (·05 c.c.) each time and another for recording automatically the evaporation of 20 mgms. of water each time are described.

Experiments are conducted to note the changes in absorption and transpiration of shoots of Barleria cristata by treating various parts of shoot with poison (formalin) to get an idea of the part

played by the living cells of the shoot in ascent of sap and it is shown that the cortical cells do not help in any way in the ascent of sap.

It is concluded that the mechanism for absorption and transpiration is not radically affected as postulated by other workers when a leaf is killed with poison.

Our thanks are due to Mr. S. Ranganathan, M.A., I.C.S., for a certain amount of preliminary work on this subject.

Reference.

1. BOSE, SIR J. C.:—Physiology of Ascent of Sap. 1921.

2. DIXON, H. H.:—Transpiration and Ascent of Sap. 1914.

3. EWART, A. J.:—The Ascent of Water in Trees. Phil. Trans. Roy. Soc. London B. 199: 341–392. 1908.

4. MACDOUGAL, D. T., OVERTON, and SMITH:—Hydrostatic-Pneumatic System in Trees. Carn. Inst. Wash. Pub. No. 397. 1929.

5. OVERTON, J. B.:—Studies on the relation of the living cells to transpiration and sap flow, in Cyperus. I. Bot. Gaz. Vol. 51. p. 28 to 63. 1911.

6.   ,,   :—Studies on the relation of the living cells to transpiration and sap flow, in Cyperus. 11. Bot. Gaz. Vol. 51. p. 102 to 120. 1911.

Table 1. Barleria cristata.

RATE OF ABSORPTION WITH THE CUT END IN WATER.

The rate indicates time taken for absorbing ·05 c.c.

Date.	Time.	Rate in minutes.	Remarks.
25–4–31	8-30 a.m.	10	
	10 ,,	$4\frac{1}{2}$	
	11 ,,	$4\frac{3}{4}$	
	12 noon.	5	
	1 p.m.	7	
	2 ,,	6	
	3 ,,	7	Fall in the rate of absorption starts.
	4 ,,	11	
	5 ,,	$12\frac{3}{4}$	
	6 ,,	$12\frac{1}{2}$	
	7 ,,	14	
	8 ,,	15	
	12 midnight	$20\frac{1}{2}$	
26–4–32	2 a.m.	,,	
	4 ,,	$17\frac{1}{2}$	Recovery in the rate of absorption begins.
	6-45 ,,	$12\frac{1}{2}$	

Table 2. Barleria cristata.

RATE OF ABSORPTION WITH THE CUT END IN 20 % FORMALIN.

The rate indicates time taken for absorbing ·05 c.c.

Date.	Time.	Rate in minutes.	Remarks.
4–10–31	9-30 a.m.	6½	cut end in water.
	10 „	5½	
	10-35 „	4½	20 % formalin supplied to the cut end.
	(Continuous	4⅓	
	readings	4½	
	between	5	
	10-30 a.m. and	5½	
	1-15 p.m.)	6	
		8	
		9	
		11	
	11-30 a.m.	13	Slight discoloration along the veins even in the top leaves.
		14	
		16	
	12-30 p.m.	27	All leaves discoloured.
		27	
	1-15 „	22	
	5-0 „	16	Leaves curling slightly.
	8-0 „	18	
	12 midnight.	23	
5–10–31	4 a.m.	22	
	7-30 „	22	

Table 3. Barleria cristata.

RATE OF ABSORPTION.

The rate indicates time taken for absorbing ·05 c.c.

Effect of coating leaves with pure formalin, with the cut end in water.

Date.	Time.	Rate in minutes.	Remarks.
6-4-31.	9-40 a.m.	$6\frac{3}{4}$	
	10-40 „	$6\frac{3}{4}$	
	11-40 „	$5\frac{3}{4}$ ⟵	Period of coating all leaves with pure formalin.
	(Continuous	6	
	readings	$6\frac{1}{2}$	
	between	7	
	11-40 a.m. &	$7\frac{3}{4}$	Excess formalin evaporated.
	1-40 p.m)	$11\frac{3}{4}$	Minimum rate of absorption.
	12-20 p.m.	$11\frac{1}{4}$	
		$10\frac{1}{2}$	
		$8\frac{1}{2}$	
		8	
		9	
		$9\frac{1}{4}$	
		$8\frac{1}{4}$	
		$7\frac{3}{4}$	
	1-40 p.m.	$7\frac{1}{2}$	Recovery in the rate is complete.
	4-40 „	$8\frac{1}{2}$	
	8-40 „	„	
	12 midnight	$10\frac{3}{4}$	
7-4-31.	6-30 a.m.	13	Leaves curled up.

Table 4. Barleria cristata.

RATE OF ABSORPTION.

The rate indicates time taken for absorbing ·05 c.c.

Effect of coating leaves with pure formalin and later on supplying the cut end with 20 % formalin.

Date.	Time.	Rate in minutes	Remarks.
22–4–31	9-20 a.m.	4	Cut end in water till 3 p.m.
	10 ,,	$5\frac{1}{4}$	
	11 ,,	$3\frac{1}{2}$	
	12 noon	$5\frac{1}{2}$	
		5 $\}$	Period of coating leaves with pure formalin.
		6	
	Continuous readings.	7	
		$7\frac{1}{2}$	Excess formalin evaporated.
		$8\frac{1}{2}$	
		9	Leaves discolouring.
		15	
	1-15 p.m.	$19\frac{1}{4}$	Minimum rate.
	3 ,, ←	9 ←	Supplied 20 % formalin to the cut end.
		9	
		$8\frac{3}{4}$	
	Continuous readings	,,	
		9	
	4 p.m.	9	
	5 ,,	$8\frac{3}{4}$	
	6 ,,	9	
	7 ,,	$9\frac{1}{2}$	
	8 ,,	10	
	12 midnight	14	
23–4–31	5-40 a.m.	15	Leaves curled up.

Table 5. Barleria cristata.

RATE OF ABSORPTION

The rate indicates time taken for absorbing ·05 c.c.

Effect of coating 6″ of the bare stem with pure formalin, with the cut end in water.

Date.	Time.	Rate in minutes.	Remarks.
5–8–31.	10-40 a.m.	10	
	11-0 ,,	$10\frac{1}{4}$	
	12 noon.	10 ←	Coated 6″ of bare stem with pure formalin.
		$10\frac{1}{4}$	
	Continuous readings.	$10\frac{1}{2}$	
		,,	No fall in the rate.
		,,	
		,,	
	1 p.m.	,,	
	2 ,,	11	
	3 ,,	,,	
	4 ,,	$10\frac{1}{2}$	
	5 ,,	$11\frac{1}{4}$	
	6 ,,	12	
	7 ,,	$12\frac{1}{2}$	
	8 ,,	,,	
	9 ,,	$13\frac{1}{2}$	

9

Table 6. Barleria cristata.

RATE OF ABSORPTION.

The rate indicates time taken for absorbing ·05 c.c.

*Effect of coating leaves first with water and later on with pure
formalin, with the cut end in water.*

Date.	Time.	Rate in minutes.	Remarks.
13–3–31.	9 a.m.	9	
	10 „	$4\frac{3}{4}$	
	11 „	$7\frac{1}{4}$	
	12 noon.	$7\frac{3}{4}$	
	12-20 p.m. ←	6 ←	Coated all leaves with water.
		$6\frac{1}{4}$	
	(Continuous	$6\frac{1}{2}$	
	readings).	10	
		13	Minimum rate.
	1 p.m.	$6\frac{3}{4}$	Excess water evaporated (leaf surface dry)
	2 „	6	
	2-35 „ ←	$6\frac{3}{4}$ ←	Coated all leaves with pure formalin.
		6	
	3 p.m.	6	
	(Continuous	$6\frac{1}{2}$	Excess formalin evaporated.
	readings	$6\frac{3}{4}$	
	from 2-35 p.m.	7	
	to 4 p.m.)	$8\frac{1}{4}$	
		$10\frac{3}{4}$	Minimum rate.
		9	
	4 p.m.	10	
	4-45 „	8	Recovery in the rate.

Table 7. Barleria cristata.

RATE OF TRANSPIRATION.

Rate indicates time taken for a loss of weight of 20 mgms.
With the cut end in water.

Date.	Time.	Rate in minutes.	Remarks.
5-2-31.	8 a.m.	$1\frac{1}{2}$	
	9 ,,	$2\frac{1}{4}$	
	10 ,,	2	Rate almost constant.
	11 ,,	2 1/3	
	12 noon	$2\frac{1}{2}$	
	1 p.m.	,,	
	1-30 ,,	3 1/5	
	2-45 ,,	3	
	3 ,,	3 4/5	Regular fall in the rate starts.
	4 ,,	4	

Table 8. Barleria cristata.

RATE OF TRANSPIRATION.

Rate indicates time taken for a loss of weight of 20 mgms.
Cut end of the shoot in 20% formalin.

Date.	Time.	Rate in minutes.	Remarks.
7–1–31.	11-55 a.m.	2	Cut end in water.
	12-15 p.m.	?	Supplied 20% formalin to the cut end.
	1-5 ,,	3	Minimum rate.
	1-50 ,,	2	Rate recovered completely; leaves discolouring.
	2-45 ,,	$1\frac{1}{5}$	Rate is quickening.
	3-45 ,,	1	Leaves completely discoloured.

Table 9. Barleria cristata.

RATE OF TRANSPIRATION.

Rate indicates time taken for a loss of weight of 20 mgms.
Coated leaves with pure formalin, with the cut end in water.

Date.	Time.	Rate in minutes.	Remarks.
17–1–31	9 a.m.	$1\frac{4}{5}$	
	10-15 ,,	$2\frac{1}{5}$	
	10-45 ,, ←	$3\frac{1}{5}$ ←	Coated leaves with pure formalin.
	11-0 ,,	$\frac{4}{5}$	Rapid rate due to evaporation of excess formalin. Leaf surface completely dry of formalin within 10 minutes.
	11-30 ,,	$1\frac{1}{2}$	Leaves getting discoloured.
	12 noon.	$1\frac{4}{5}$	
	1 p.m.	2	Leaves curling.
	2 ,,	3	Fall in the rate.
	3 ,,	$3\frac{1}{2}$	
	4-30 p.m.	$5\frac{3}{5}$	Leaves shrivelled up.

Table 10. Barleria cristata.

RATE OF TRANSPIRATION.

Rate indicates time taken for a loss of weight of 20 mgms.
Water supply at the cut end withdrawn.

Date.	Time.	Rate in minutes.	Remarks.
30-1-31.	8-30 a.m.	$2\frac{3}{4}$	Cut end in water.
	9-45 ,,	$1\frac{3}{4}$	Rate is fairly constant.
	10-0 ,,	$1\frac{1}{2}$ a.m	Water supply at the cut end is withdrawn.
	11 ,, ←	3·6 ←	Leaves are drooping.
	12 noon.	6	
	12-45 p.m.	$6\frac{1}{2}$	All leaves completely drooping.

Table 11. Barleria cristata

CHANGES IN THE SIZE OF THE STOMATAL APERTURES.

Water sucked up in the porometers each time to 8 cms. and the time taken for a fall of ·5 cm. noted,

Date.	Time.	CONTROL. (POROMETER) Time taken for a fall of ·5 cm.	Remarks.	EXPERIMENTAL ONE (POROMETER) Time taken for a fall of ·5 cm.	Remarks.
		min. sec.		min. sec.	
1-9-31	8-30 a.m.	10 - 40		5 - 0	
	9 ,,	2 - 40	Just after cutting	4 - 40	
	9-30 ,,	3 - 0		4 - 35	
	9 ,,	5 - 35		4 - 55 ←	20% formalin supplied to the cut end and at 10-13 a.m.
	10-15 ,,	7 - 25		4 - 15	
	10-30 ,,	5 - 30		5 - 5	No change in the stomata
	10-45 ,,	4 - 40		4 - 35	
	11 ,,	3 - 35		4 - 50	
	11-30 ,,	4 - 10		4 - 55	
	12 noon	5 - 30		5 - 10	
	12-30 p.m.	11 - 20		6 - 5	Veins of expt. leaf discoloured.
	1 ,,	12 - 30	Stomata closing	5 - 5	
	1-30 ,,	11 - 40		4 - 20	
	2-30 ,,	8 - 40		3 - 0	All leaves discoloured.
	3-30 ,,	10 - 55		2 - 50	
	4-45 ,,	6 - 25	Direct sunlight	3 - 25	Stomata comparatively wide open